

San Luis & Delta-Mendota Water Authority



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April 25, 2014

Delivered Via E-mail to kari.holmes@waterboards.ca.gov

Ms. Kari Holmes
California Regional Water Quality Control Board
Central Valley Region
11020 Sun Center Drive, Suite 200
Rancho Cordova, CA 95670

Subject: Comments on the Tentative Order R5-2014-XXXX, NPDES No. CA0079138, for the City of Stockton Regional Wastewater Control Facility, San Joaquin County

Dear Ms. Holmes:

The State Water Contractors (SWC) and San Luis & Delta-Mendota Water Authority (Authority)¹ (collectively, "Public Water Agencies") appreciate the opportunity to provide comments on the Central Valley Regional Water Quality Control Board's (Regional Water Board) Tentative Renewal of Waste Discharge Requirements (NPDES No. CA0079138) for the City of Stockton Regional Wastewater Control Facility (tentative permit). The Public Water Agencies have a significant interest in the health of the San Francisco Bay/Sacramento-San Joaquin River Delta (Bay-Delta) ecosystem and in the development of management actions to address stressors on the Bay-Delta, including water quality impairment. With respect to municipal wastewater discharges, we support the development and implementation of management actions to address nutrient impacts in the Bay-Delta ecosystem.

The Public Water Agencies commend the Regional Water Board staff for their commitment to protect municipal and aquatic life beneficial uses, including both in the vicinity of the City of Stockton's Regional Wastewater Control Facility (Stockton RWCF) discharge and downstream, and for their efforts to address nutrient discharges to the Bay-Delta through the development of the tentative permit. The Public Water Agencies support the tentative permit, and in particular support the revised ammonia limits and the more stringent nitrate plus nitrite limit of 10 mg/L. Excess nutrient loading to the Bay-Delta can contribute to excess and/or nuisance algae blooms that result in significant impacts including ecosystem changes, depressed dissolved oxygen, cyanotoxin production, nuisance to recreational uses and water provider operations, and taste and odor issues for drinking water supplies. As the State Water Resources Control Board (SWRCB) found when upholding the discharge permit issued to the Sacramento Regional County Sanitation District, "The total amount, or load, of nitrogen needs to be reduced in the Delta in order to address the damaging effects of nutrient over-enrichment. Since ammonia and

¹ For a description of the SWC and San Luis & Delta-Mendota Water Authority see Attachment 1.

nitrate are the dominant forms of nitrogen from point source discharges, the loads of both forms of nitrogen to waterbodies experiencing excessive biostimulation needs to be reduced. Elevated levels of ammonia are toxic and thus the conversion to nitrate through nitrification is necessary to protect aquatic life beneficial uses. However, converting the dominant form from primarily ammonia to nitrate will still result in cultural eutrophication. Reductions of total nitrogen loads through both nitrification and denitrification is the goal to protect beneficial uses from cultural eutrophication from point source discharges.” (SWRCB Order WQ 2012-0013 at 30).

The Public Water Agencies do not support the Nitrate Option for the tentative permit. The Nitrate Option is based solely on protecting the municipal and domestic water supply (MUN) beneficial use and further narrowly interprets protecting the MUN beneficial use as meeting the California Department of Public Health maximum contaminant level (MCL) for nitrate plus nitrite of 10 mg/L (as N) assuming a mixing zone. This option does not address the on-going impacts of nitrate loading on aquatic life beneficial uses, and does not address compliance with the narrative objectives for biostimulatory substances and tastes and odors. The discharge of nitrate from the Stockton RWCF contributes to nutrient over-enrichment of the San Joaquin River and Delta, and impacts aquatic life and MUN beneficial uses. The attached comments on the tentative permit (Attachment 2) provide additional evidence to support the 10 mg/L nitrate plus nitrite limit.

The Public Water Agencies also reviewed the City of Stockton’s report “Evaluation of the Potential Effects of Nitrate Plus Nitrite Discharged from the Stockton Regional Wastewater Control Facility on the San Joaquin River in Support of Dilution Credit for NPDES Permitting” (Nitrate Study). Comments on the Nitrate Study are also included in Attachment 2. Further evidence of nutrient impacts in the Bay-Delta is provided in the attached Nutrient Science Summary Technical Memorandum (Attachment 3) that documents nutrient impacts on aquatic life in the Bay-Delta Estuary, and includes numerous referenced studies concerning the impacts of nutrient loading on the Bay-Delta ecosystem.

We appreciate the opportunity to provide comments on the tentative permit and look forward to continuing to work with the Regional Water Board on the permit renewal process. If you have any questions concerning these comments, please contact Lynda Smith, Metropolitan Water District of Southern California, at 916-650-2632 or lsmith@mwadh2o.com.

Sincerely,



Daniel G. Nelson
Executive Director
San Luis & Delta-Mendota Water Authority



Terry L. Erlewine
General Manager
State Water Contractors

Attachments

Attachment 1

San Luis & Delta-Mendota Water Authority: The San Luis & Delta Mendota Water Authority (Authority) is a joint powers authority, established under California's Joint Exercise of Powers Act. (Gov. Code, § 6500 et seq.). The Authority is comprised of 28 member agencies. The Authority member agencies have historically received up to 3,100,000 acre-feet annually of Central Valley Project (CVP) water for the irrigation of highly productive farm land primarily along the San Joaquin Valley's Westside, for municipal and industrial uses, including within California's Silicon Valley, and for publicly and privately managed wetlands situated in the Pacific Flyway. The areas served by the Authority's member agencies span portions of seven counties encompassing about 3,300 square miles, an area roughly the size of Rhode Island and Delaware combined. The Authority's members are: Banta-Carbona Irrigation District; Broadview Water District; Byron Bethany Irrigation District (CVPSA); Central California Irrigation District; City of Tracy; Del Puerto Water District; Eagle Field Water District; Firebaugh Canal Water District; Fresno Slough Water District; Grassland Water District; Henry Miller Reclamation District #2131; James Irrigation District; Laguna Water District; Mercy Springs Water District; Oro Loma Water District; Pacheco Water District; Panoche Water District; Patterson Irrigation District; Pleasant Valley Water District; Reclamation District 1606; San Benito County Water District; San Luis Water District; Santa Clara Valley Water District; Tranquillity Irrigation District; Turner Island Water District; West Side Irrigation District; West Stanislaus Irrigation District; Westlands Water District.

State Water Contractors, Inc. (SWC): The SWC organization is a nonprofit mutual benefit corporation that represents and protects the common interests of its 27 member public agencies in the vital water supplies provided by California's State Water Project ("SWP"). Each of the member agencies of the State Water Contractors holds a contract with the California Department of Water Resources ("DWR") to receive water supplies from the SWP. Collectively, the SWC members deliver water to more than 25 million residents throughout the state and more than 750,000 acres of agricultural lands. SWP water is served from the San Francisco Bay Area, to the San Joaquin Valley and the Central Coast, to Southern California. The SWC's members are: Alameda County Flood Control and Water Conservation District Zone 7; Alameda County Water District; Antelope Valley-East Kern Water Agency; Casitas Municipal Water District; Castaic Lake Water Agency; Central Coastal Water Authority; City of Yuba City; Coachella Valley Water District; County of Kings; Crestline-Lake Arrowhead Water Agency; Desert Water Agency; Dudley Ridge Water District; Empire-West Side Irrigation District; Kern County Water Agency; Littlerock Creek Irrigation District; Metropolitan Water District of Southern California; Mojave Water Agency; Napa County Flood Control and Water Conservation District; Oak Flat Water District; Palmdale Water District; San Bernardino Valley Municipal Water District; San Gabriel Valley Municipal Water District; San Geronio Pass Water Agency; San Luis Obispo County Flood Control & Water Conservation District; Santa Clara Valley Water District; Solano County Water Agency; and Tulare Lake Basin Water Storage District.

Attachment 2

Public Water Agencies Comments on the Tentative Order for the City of Stockton Regional Wastewater Control Facility

The Public Water Agencies commend the Regional Water Board staff for their commitment to protect municipal and aquatic life beneficial uses, including both in the vicinity of the City of Stockton's Regional Wastewater Control Facility (Stockton RWCF) discharge and in downstream waterbodies and for their efforts to address nutrient discharges to the Bay-Delta through the development of the tentative permit. The Public Water Agencies support the tentative permit, and in particular support the revised ammonia limits and the more stringent nitrate plus nitrite limit of 10 mg/L.

In the tentative permit, the Regional Water Board properly finds that the contribution of nitrate from the Stockton RWCF can contribute to excessive algal growth and change the ecology of Delta waterbodies, which has impacts on aquatic life and municipal beneficial uses. The Regional Water Board also properly finds that the discharge of nitrate from the Stockton RWCF has reasonable potential to cause or contribute to an exceedance of the Basin Plan narrative water quality objectives for biostimulatory substances and taste and odors. The applicable narrative water quality objectives are the following:

- Biostimulatory Substances. Water shall not contain biostimulatory substances which promote aquatic growths in concentrations that cause nuisance or adversely affect beneficial uses.
- Taste and Odors. Water shall not contain taste- or odor-producing substances in concentrations that impart undesirable tastes or odors to domestic or municipal water supplies or to fish flesh or other edible products or aquatic origin, or that cause nuisance, or otherwise adversely affect beneficial uses.

Further, the Regional Water Board properly finds that excessive algae growth and the resulting impacts on downstream waterbodies and municipal drinking water supplies (i.e., increased organic carbon, filter clogging algae, and taste and odor producing algae) are occurring and that nutrient loading contributes to the impairment of beneficial uses. Finally, the Regional Water Board properly established the effluent limit for nitrate plus nitrite at 10 mg/L (as N), based on the technical capability of wastewater treatment plants to achieve the limit using standard denitrification technologies.

The following Public Water Agencies' comments on the tentative permit and the City of Stockton (Discharger) report on "Evaluation of the Potential Effects of Nitrate Plus Nitrite Discharged from the Stockton Regional Wastewater Control Facility on the San Joaquin River in Support of Dilution Credit for NPDES Permitting" (Nitrate Study) provide additional evidence to support the findings in the tentative permit.

Impacts of Nutrients on Drinking Water Supplies

The Public Water Agencies and their member agencies who take water from the State Water Project (SWP) and Central Valley Project (CVP) are already challenged by the algae and macrophyte growth that occurs in the SWP and CVP aqueducts and reservoirs. The challenges include filter-clogging algae, taste and odor producing algae, and aquatic macrophytes that clog

conveyance structures and cause numerous other problems. The Stockton Nitrate Study hypothesizes that, “When waters of the Delta are exported into relatively shallow conveyance canals, algae *may* (emphasis added) no longer be light limited, and growth of epibenthic algae and submerged aquatic vegetation *may* (emphasis added) occur, in addition to phytoplankton growth.” In fact, the problems with algal and aquatic plant growth in the SWP system are well documented (Archibald Consulting et al. 2012, Department of Water Resources 2013). Algal and aquatic plant growth in the SWP conveyance facilities and downstream reservoirs is neither light-limited nor inhibited by high ammonium concentrations since most of the ammonium from the Delta has been nitrified to nitrate. Elevated levels of nutrients (phosphorus and nitrogen compounds) stimulate nuisance algal and aquatic weed growth that includes production, by specific cyanobacteria, of noxious taste and odor (T&O) compounds and algal toxins. In addition to algal produced T&O and algal toxin concerns, increases in algal and aquatic weed biomass can impede flow in conveyances, shorten filter run times and increase solids production at drinking water treatment plants, and add to organic carbon loading.

The Discharger’s Nitrate Study includes results of a Delta hydrologic modeling study that evaluated the proportion of water at the SWP, CVP and other drinking water intakes that is Stockton RWCF effluent, and the incremental contribution of nitrate from Stockton RWCF at the drinking water intakes, assuming the permitted flow of 55 million gallons per day (MGD) and different options for the nitrate limit. The study results indicate that the Stockton RWCF discharge would increase nitrate concentrations at the SWP Banks Pumping Plant by 0.29 mg/L-N on a long-term average and up to 1.0 mg/L-N as a daily maximum, assuming the Discharger proposed seasonal nitrate limit option of 26 mg/L-N April to September and 30 mg/L-N October to March. The corresponding increase at the CVP Jones Pumping Plant is 0.20 mg/L-N on a long-term average and up to 0.91 mg/L-N as a daily maximum. The Public Water Agencies used these modeling results and the historical nitrate data for the SWP Banks Pumping Plant to analyze the incremental increase in nitrate at Banks Pumping Plant assuming a Stockton RWCF discharge of 55 MGD and seasonal nitrate limits of 26/30 mg/L-N. This analysis shows that on average the Stockton RWCF discharge would increase nitrate concentrations at the Banks Pumping Plant by 12% to 98% (see **Table 1**). The largest increase in nitrate concentrations would occur during the summer months when algal problems already occur in the SWP.

Taste and Odor Problems

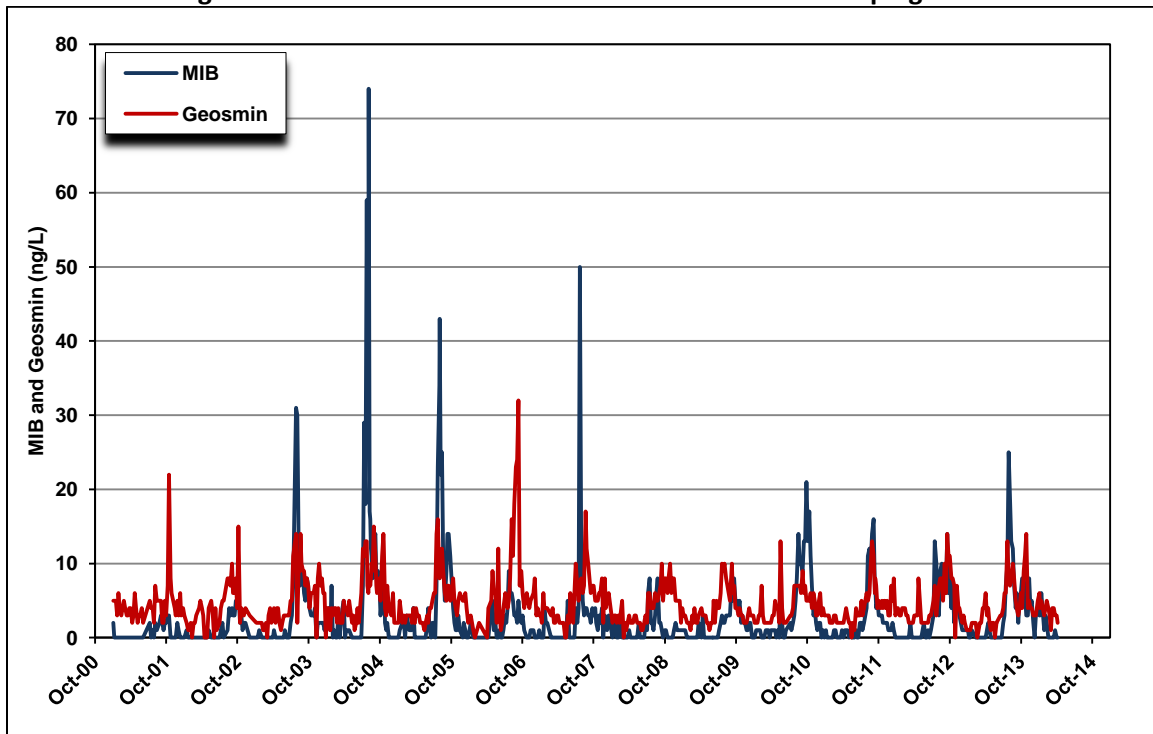
Certain cyanobacteria and actinomycete bacteria produce chemical compounds that are not removed in conventional water treatment processes and are capable of causing unpleasant tastes and odors in drinking water. T&O incidents occur throughout the SWP and SWP, and are commonly associated with geosmin and MIB that are produced by benthic and planktonic cyanobacteria. The ability of individuals to detect these chemicals varies, but the general population can detect either compound at a concentration of about 10 ng/L (parts per trillion) and sensitive individuals can detect even lower concentrations.

Table 1. Incremental Increase in Nitrate Concentrations at Banks Pumping Plant Due to Stockton RWCF Discharge at permitted level of 55 MGD

Month	Average Nitrate at Banks PP (mg/L) ^a	Stockton Incremental Long-term Average Contribution (mg/L)	Average Nitrate at Banks PP with Stockton Incremental Contribution (mg/L)	Percent Increase from Stockton RWCF Discharge ^b
Jan	1.27	0.29	1.56	12%
Feb	1.15	0.29	1.44	14%
Mar	1.00	0.29	1.29	16%
Apr	0.65	0.29	0.94	24%
May	0.48	0.29	0.77	33%
Jun	0.49	0.29	0.78	32%
Jul	0.29	0.29	0.58	54%
Aug	0.16	0.29	0.45	98%
Sep	0.18	0.29	0.47	86%
Oct	0.30	0.29	0.59	53%
Nov	0.60	0.29	0.89	26%
Dec	0.81	0.29	1.10	20%
<p>a. Source of nitrate data: California Data Exchange Center (CDEC) station HRO and the Department of Water Resources Water Data Library station KA000331. The period of record is 1968 to 1979 and 1996 to 2013.</p> <p>b. Stockton RWCF's current discharge of about 26 MGD contributes nitrate to Banks PP, so this analysis of the incremental increase in nitrate contribution is based on the permitted increase of 29 MGD in effluent flow up to 55 MGD.</p>				

Samples have been collected from SWP facilities and analyzed for the T&O producing compounds, MIB and geosmin, since 2000. **Figure 1** shows that peak concentrations of MIB and geosmin occur each summer at the Banks Pumping Plant and levels exceeding 10 ng/L have been present for a number of weeks each summer in most years. Benthic cyanobacteria are the primary sources of T&O compounds in the Delta and in Clifton Court Forebay. The high levels of MIB and geosmin are transported to the South Bay Aqueduct (SBA) and down the California Aqueduct. MIB and geosmin are also generated by benthic cyanobacteria in the California Aqueduct, the Coastal Branch and the East Branch of the California Aqueduct. MIB and geosmin are both frequently present at high concentrations in the East Branch of the aqueduct. The maximum concentrations recorded were 240 ng/L of MIB in May 2003 and 396 ng/L of geosmin in July 2012. Planktonic cyanobacteria are responsible for T&O problems in Silverwood Lake, Lake Perris, Pyramid Lake, Castaic Lake, and Lake Skinner in southern California. The Department of Water Resources (DWR) uses copper compounds in the SWP aqueducts and reservoirs to control these cyanobacteria, as does the Metropolitan Water District of Southern California in its reservoirs that store SWP supplies. A hypolimnetic aeration system was installed in Lake Perris but it is not able to effectively control algal blooms due to the excessive nutrient load to the lake.

Figure 1. MIB and Geosmin Concentrations at Banks Pumping Plant



Algal Toxins

Microcystis aeruginosa was first detected in the Delta in the eastern Stockton Ship Channel on September 27, 1999. This cyanobacteria has bloomed every year following its initial detection during the late summer and early fall throughout the central and southern Delta. DWR initiated cyanotoxin monitoring in the SWP in 2006. Microcystin-LR was detected at all of the locations that were monitored in 2007, except Barker Slough. Although it was detected, it was below the reportable limit of 1 µg/L so the concentrations could not be quantified. Microcystin-LA was detected at the Pacheco Pumping Plant and the outlet tower of San Luis Reservoir in 2012. The concentrations at the Pacheco Pumping Plant ranged from < 1.0 to 1.6 µg/L. The outlet tower concentrations ranged from < 1.0 to 1.7 µg/L. A trace amount of microcystin-LR was detected in one sample from the outlet tower. There are currently no state or federal drinking water standards for microcystins; however, the World Health Organization released a provisional guideline of 1.0 µg/L for microcystin-LR in drinking water in 1998. The United States Environmental Protection Agency (USEPA) added cyanobacteria and cyanotoxins to the candidate contaminant list (CCL) IN 1998, 2005 and 2009.

Filter Clogging Algae

Filter clogging algae occur throughout the SWP but they are particularly troublesome in the SBA. The high concentrations of nutrients, combined with shallow canal depth, abundant sunlight, and warm water temperatures during the spring, summer, and fall months leads to excessive algal growth in the SBA. This creates a number of treatment challenges for the SBA Contractors. A benthic diatom, *Melosira* sp., forms chains of cells that are sloughed off of the bottom when the chains become long and this leads to filter clogging problems at SBA water treatment plants. The population of *Melosira* generally increases from March to July and then again in the fall months (Personal Communication, Jeff Janik, DWR). The primary mechanism

for controlling algal growth in the SBA is by application of copper sulfate. Copper sulfate is applied from March or April until September, depending upon water temperatures and algal conditions. Copper sulfate effectively reduces algal populations but the dead algae release T&O producing compounds.

Conveyance Impacts

Excessive growth of aquatic weeds and algae create water conveyance problems at a number of locations in the SWP and CVP facilities. Aquatic weed accumulation can be so severe at the Banks Pumping Plant that pumping is restricted or halted. At times, up to 20 cubic yards of aquatic weeds are removed each day from the trash racks at the Banks Pumping Plant. Aquatic weeds also create major operational problems in O'Neill Forebay, the California Aqueduct and the Coastal Branch. Aquatic weeds are also present in the littoral zone of the four southern California SWP reservoirs. DWR expends a significant amount of time and money controlling aquatic weeds in the SWP. Copper products are used in many locations, although they have not been used since 2006 in Clifton Court Forebay due to potential impacts on listed species. Mechanical harvesting is used in Clifton Court Forebay and O'Neill Forebay and some sections of the aqueduct are scraped by dragging a large chain along the aqueduct lining.

Solids Production

Water agencies must use additional quantities of chemicals in the water treatment process to remove algae from the source water. This produces greater quantities of solids that must be disposed, resulting in higher disposal costs.

Increased Organic Carbon

Algal production in the SWP facilities results in higher concentrations of total organic carbon in the system. The amount of organic carbon that must be removed by a water treatment plant is based on the concentrations of total organic carbon and alkalinity in the source water.

Comments on the City of Stockton report on "Evaluation of the Potential Effects of Nitrate plus Nitrite Discharged from the Stockton Regional Wastewater Control Facility on the San Joaquin River in Support of Dilution Credit for NPDES Permitting" (Nitrate Study)

The Nitrate Study report prepared by Robertson-Bryan, Inc., describes the Discharger's current understanding of, and recent data collection results and modeling efforts, related to nutrient discharge from the Stockton RWCF and its potential impacts. The study correctly addresses a number of key facts about nutrients in the San Joaquin River and the discharge of the Stockton RWCF. These include:

- The San Joaquin River is a very nutrient rich river, and with respect to nitrogen, the bulk of this is in the form of nitrate, NO₃.
- The nutrient character of the San Joaquin River differs substantially from that of the Sacramento River, where the bulk of the nitrogen discharge in the form of wastewater effluent is as ammonium, NH₄.
- There are multiple nutrient sources on the San Joaquin, and these, as well, differ from those of the Sacramento River.
- Nutrient levels in the San Joaquin River have been increasing over time.

- Recognition that nitrate concentrations and abundance of *Microcystis* rarely correlate. (Note, this is because nitrate is not a preferred form of nitrogen for *Microcystis*, a point not recognized in the report).

The Nitrate Study attempts to make the case that nutrient loads that are high are not a significant problem and therefore not a reason to deny mixing zones. They base this conclusion on a number of questionable assumptions, limited recognition of or reference to the broader literature, limited data and several specific errors. Each of these points is addressed below.

The first assumption that the authors present in defense of their case that nutrients are not a concern is the notion that since nutrients are in excess they are not limiting. If they are not limiting then some other factor, typically light, must be the controlling factor. They cite historical data from the Delta and modeled results from Jassby suggesting this is the case. However, such a notion fails to recognize that even in excess nutrients play important regulatory roles in ecosystem structuring. Nutrient loads set the total biomass that can be supported in an ecosystem. Nutrients in both limitation and in excess may affect cell metabolism and chemical composition (e.g., Glibert et al. 2013). The latter, in turn has effects on the food web at all levels. Moreover, the authors fail to acknowledge that new results suggest that the historic modeled primary production measurements of Jassby may have changed over time (Parker et al., 2012). The role of light as a limiting factor is, of course, not dismissed as a potential factor regulating growth during some times, but general statements about light being always limiting have not been substantiated. Photoacclimation and photoadaptation are not merely responses to absolute irradiance, but depend on the balance of nutrient and energy flow through the cell, as well as inherent differences between different types of algae (Kana et al., 1997, MacIntyre et al. 2002). As algae have changed through the years, as nutrients have changed through the years, it is imperative that old assumptions be revisited.

Nutrients in excess affect many aspects of cell metabolism. As fully detailed in Glibert et al. (2013), growth is a dynamic balance between assimilative processes and dissipative processes, with varying emergent properties changing in response to nutrient levels – even when these nutrients are not at limiting concentrations. Such properties include the relative proportions of different enzymes, gene regulation, cell pigmentation and cell composition, both in terms of nutrient proportions and lipid. As well documented in the literature on ecological stoichiometry (a literature that was entirely ignored by the authors of this report), the relative properties of prey (including the algae) have important ecological consequences for their predators (e.g. Sterner and Elser 2002). The literature on ecological stoichiometry, the balance of energy and multiple elements in ecological interactions and processes, conceptualizes the relative balance of nutrients affects all aspects of behavior, reproduction, fecundity, and ultimately their growth and success (e.g., Jeyasingh and Weidner, 2005, 2007). Grazers are affected by the food quality available to them, and they, in turn, affect the food web by altering the composition of food available to their prey (e.g., Sterner and Elser 2002, De Troch et al. 2012). Additional specific misconceptions of the authors with regard to ecological stoichiometry are noted in more detail below.

The authors of the Nitrate Study fail to recognize that when nutrients are not assimilated in a given region – i.e., in the limited region over which they made their measurements, the nutrients are exported downstream where they have the potential to contribute to eutrophication or other effects in regions displaced in space or time from the specific site and time of discharge. That the nutrients are not assimilated in the immediate zone of mixing

should not be reason to assume they do not contribute to effects elsewhere. The only possible fates of these nutrients are displacement downstream, volatilization to the atmosphere, seepage into groundwater or denitrification. Given the dominant form of nitrogen (N) is nitrate, volatilization to the atmosphere is small. Rates of denitrification measured in the south eastern Delta, suggest that while substantial, denitrification rates are not large enough to remove more than a small percent of the ambient nitrate (Cornwell et al. 2014). The authors have limited the regional scope of their study to immediate zones of influence and have failed to consider the export of these nutrients downstream and beyond the SWP and CVP pumping plants.

The authors of the Nitrate Study also assume that more nutrients make no difference when concentrations are already above saturation levels. The first problem with this assumption is they base their argument for saturation on a very limited set of numbers of half saturation constants (p. 7) from the older literature. They fail to recognize that half saturation constants are highly variable depending on the type of cell (e.g., algal taxa), its chemical composition and physiological state among other factors. For example, some algae, especially diatoms, continue to take up nitrate well in excess of saturation parameters (e.g., Collos et al., 1992; Watt et al., 1992; Collos et al., 1997; Lomas and Glibert, 1999a,b). Second, nutrients at concentration levels beyond saturation do matter for algal metabolism and can affect important cell constituents. For example, many toxic algae, a concern raised in this report, and that show an increase in abundance in the Bay-Delta, produce toxins in greater amounts when growing under a nutrient regime that is not in stoichiometric proportion (Flynn et al. 1994, 1995; Johansson and Granéli 1999a,b; Granéli and Flynn, 2006). Many cyanobacteria, for example, produce more toxins when growing at high N:P ratios (e.g., Lee et al., 2000; Vézic et al., 2002; Downing et al., 2005; Lehman 2007, Ha et al. 2009; Van de Waal et al., 2009); many of these toxins are rich in N. Even if there were no other points of contention in this document, the fact that cyanobacteria may be more toxic at higher N:P ratios should give pause to any consideration that changes in nutrient ratios do not matter.

Third, the Nitrate Study concludes that the incremental increase in nitrate concentrations from the Stockton RWCF at the Banks and Jones pumping plants will not cause algal blooms or result in undesirable tastes and odors for downstream water users when they otherwise would not occur. However, there is no evidence to support the conclusion that adding additional nitrate from the Stockton RWCF as average dry weather effluent flow increases to 55 MGD would not exacerbate these problems. The authors make the calculation (see Nitrate Study, Table 9) purporting to illustrate that the incremental nitrate addition at Delta water intakes would be small regardless of whether the effluent concentrations were at 10 or at 26/30 mg/L-N. These concentrations can be viewed from a different perspective if one recalculates these values in the units of $\mu\text{M-N}$ and then applies a well-documented (and conservatively estimated) relationship of chlorophyll-a biomass resulting from new N additions as 1 $\mu\text{g/L chl}$ for each 1 $\mu\text{M-N}$ (e.g., McCarthy et al. 1977). The table has been reproduced here (**Table 2**), illustrating the potential incremental increase in chlorophyll (assuming no loss to denitrification or other pathways). As can be seen, under many of the conditions such an incremental addition can shift the downstream water into a condition of at least mesotrophy, if not eutrophy, as defined using the authors own criteria (based on EPA values and as reported in Table 1, Nitrate Study).

Table 2. Potential Incremental Increase in Chlorophyll as a Result of Increased Nitrate Concentrations

Location	Effluent Limit of 10 mg/L as N			Effluent Limit of 26/30 mg/L as N		
	Long-term Ave. Nitrate (mg/L) In parens are uM values	Daily max (mg/L) In parens are uM values	Potential new downstream chl a (ug/L)	Long-term ave (mg/L) In parens are uM values	Daily max (mg/L) In parens are uM values	Potential new Downstream chl a (ug/L)
Barker Slough	0.0 (0.0)	0.0 (0.0)	0.0	0.0 (0.0)	0.0 (0.0)	0.0
Sac at Mallard Isl	0.0 (0.0)	0.04 (2.86)	0.0-2.86	0.01 (0.71)	0.10 (7.14)	0.71-7.1
San J at Antioch	0.01 (0.71)	0.07 (5.0)	0.71-5.0	0.02 (1.42)	0.18 (12.9)	1.4-12.9
San J at Venice	0.02 (1.42)	0.25 (17.9)	1.4-17.9	0.06 (4.29)	0.65 (46.4)	4.3-46.4
CCWD Rock Slough PP	0.02 (1.42)	0.19 (13.6)	1.4-13.6	0.06 (4.29)	0.51 (36.4)	4.3-36.4
Victoria Canal	0.18 (12.9)	0.66 (47.1)	12.9-47.1	0.50 (35.7)	1.99 (142.1)	35.7-142
Cliffton Court Forebay	0.10 (7.14)	0.34 (24.3)	7.1-24.3	0.29 (20.7)	1.02 (72.9)	20.7-72.9
Delta Mendota Canal	0.07 (5.0)	0.34 (24.3)	5.0-24.3	0.20 (14.3)	0.91 (65.0)	14.3-65.0

There are several areas of concern in the Nitrate Study regarding the specific methodology used for sampling. Algae samples were collected and preserved with glutaraldehyde and the bottles were “shaken vigorously five times” (p. 24). Such an approach would compromise any fragile and delicate species. Thus, it must be assumed that such species are underrepresented in these samples. The algal data also do not include any picoplankton. No replicates, standard deviations, standard errors or lower limits of detection were reported for the nutrient constituents. Additionally, estimates of coverage of macrophytes were made by field crew who “wore polarized glasses to maximize their ability to see below the surface.” This is a highly unusual and subjective approach at best, and there is no evidence that any of these measurements were quantitative. It would appear that these were no more than guesses. Finally, all measurements were apparently made during mid-day; thus no estimates are provided of potential for diel dissolved oxygen sags.

A number of errors, misconceptions or inconsistencies were also apparent in the report.

- 1) Nitrate Study page 11: The report states that there is no evidence to indicate that a modest increase in the nitrogen to phosphorus (N:P) ratio in the range of 10:1 to 40:1 would have any significant effect on the abundance of *Microcystis* in the Delta. A 4-fold change in N:P (using their estimated range of 10:1 to 40:1) is not a “modest” change.
- 2) The authors have misrepresented the debate over the Glibert (2010) analysis. While the authors claim that the statistical approach was flawed and therefore the results are invalid, this is not correct. Yes, the use and reporting of the “p” value was incorrect and appropriately noted in the literature, the use of the overall statistical technique, CUSUM, is one of the most powerful approaches in trend analysis. The conclusions of that paper have since been reaffirmed using other approaches (Glibert et al., 2011).

- 3) Statistical results were based on very limited sampling and a small range in nutrient parameters.

There are also many statements that are unclear. As an example, on page 10, it is stated, “Although the high nutrient (nitrogen and phosphorus) concentrations found throughout the Delta were cited as prerequisites for the bloom, the persistence of the bloom was not related to nutrients because nutrient concentrations are much higher than limiting values throughout the Delta (Lehman et al. 2008).” What the authors are trying to convey is a bit of twisted logic, in that they are saying that because the nutrients were not in limiting values then nutrients are not related to the bloom. However, if nutrients were limiting there would be no bloom- and no persistence of the bloom – limiting nutrients are just that- limiting. When nutrients are at limiting concentrations they limit growth. That is, nutrients that are required for growth cannot be regulating persistence of a bloom by their absence. This is just one of many examples where the concept of limiting nutrient has been over-interpreted or misinterpreted throughout this document. Indeed, other factors can be related to bloom persistence, but lack of nutrients is absolutely not one of them.

The correlations provided in the Nitrate Study with regard to algal communities and various nutrient parameters are interesting, but very limited. The range of variation in the nutrients overall was small (as was the total number of observations), so lack of relationships may simply be a function of the size of data set. Virtually no observations in the data set reported are of values comparable to those observed in the southeastern Bay-Delta in prior decades, as shown in Figures 2 and 3 below. Long term data from the eastern and southeastern regions of the Delta (region encompassing south of Stockton to Franks Tract) show increasing trends in N:P ratio over time (data in molar units). There is strong support in the scientific literature for the proposition that the N:P ratio influences phytoplankton community composition. Attachment 3 includes additional discussion on the significance of changes in N:P ratios in the Delta.

Figure 2. Changes in N:P Ratio in the spring months (March to June) over time for the eastern and southeastern regions of the Delta.

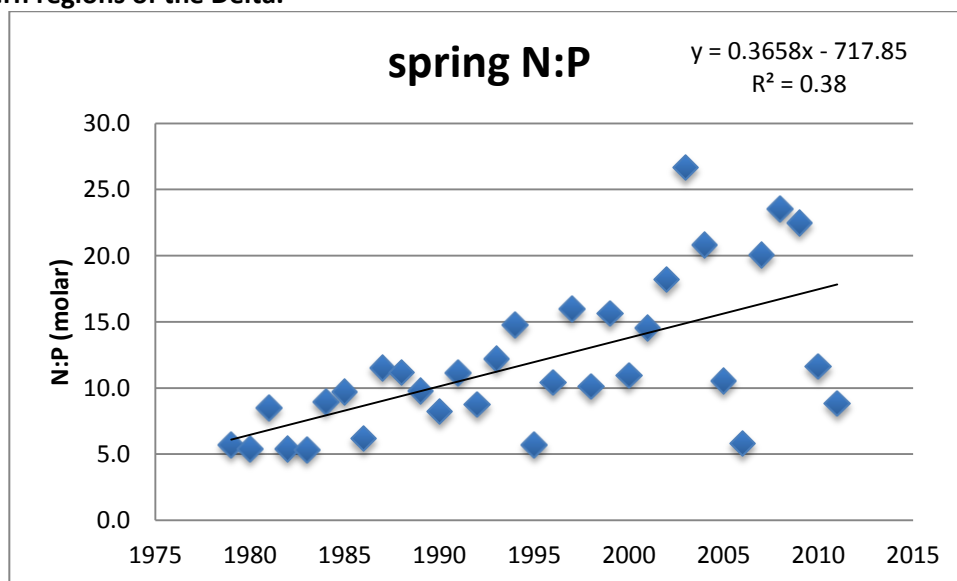
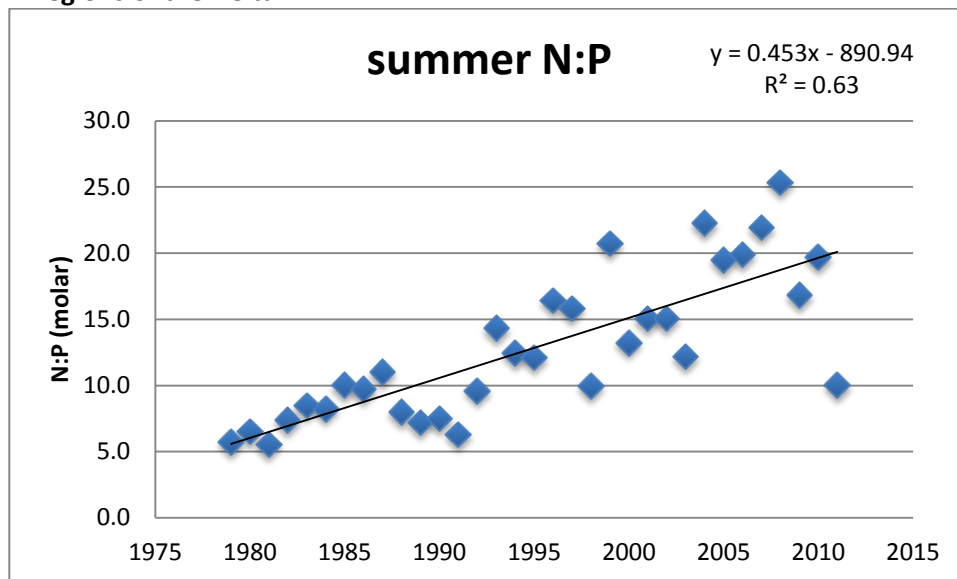


Figure 3. Changes in N:P Ratio in the summer months (July to October) over time for the eastern and southeastern regions of the Delta.



Mixing Zone

The above technical analysis provides further support to the Regional Board's reasoned proposal to deny a mixing zone for nitrate plus nitrite. Indeed, the proposal is fully consistent with established agency guidance and should be maintained in the final permit.

It is a fundamental requirement of the Clean Water Act that a discharger must meet all permitted effluent limits at the "end of pipe" – the point where the pollution leaves a plant and enters the receiving waters of the State and/or the United States. By regulation, U.S. EPA allows state permitting agencies the discretion to create a limited exception known as a "mixing zone" that would allow a discharger to exceed a limit at the discharge point and dilute or "mix" the pollution in the receiving water. However, U.S. EPA has urged that this exception be applied "carefully" because of the additional loadings resulting from a mixing zone. EPA Water Quality Standards Handbook § 5.1 ("Mixing zone allowances will increase the mass loadings of the pollutant to the water body and decrease treatment requirements. . . . Because of these and other factors, mixing zones must be applied carefully so as not to impede progress toward the Clean Water Act goals of maintaining and improving water quality.")² EPA also urged that a mixing zone should not be permitted unless the state agency has "determined that the assimilative capacity of the receiving system can safely accommodate the discharge. . . . [taking] into consideration the physical, chemical, and biological characteristics of the discharge and the receiving system; the life history and behavior of organisms in the receiving system; and the desired uses of the waters." *Id.*

The State Board has allowed a mixing zone exception – but it is a very narrow one – and committed to the informed discretion of the Regional Board. Under the Basin Plan, a Regional Board may deny a request for a mixing zone if the discharger has not "demonstrated to the satisfaction of the Regional Water Board that the mixing zone will not adversely impact

² Available at <http://water.epa.gov/scitech/swguidance/standards/handbook/chapter05.cfm>

beneficial uses.” (Basin Plan, at IV-16.00.). As such, the State Board has repeatedly upheld a regional board’s broad discretion to deny a request for mixing zones and dilution credits. *E.g.*, *In the Matter of the Petition of Yuba City*, WQO 2004 – 0013, 10 (July 22, 2004) (citing cases, holding the “regional boards have discretion in allowing mixing zones and dilution credits...”); *In the Matter of the Petition of City of Stockton*, WQO 2003 – 0002, 3 (March 19, 2003) (citing consideration of “real-time flow data . . . which shows that dilution has been minimal during above-average wet years for all conditions,” as well as “inadequacy of existing models, the fact that the receiving water is impaired, and the presence of threatened and endangered species”). So long as the Regional Board considered the available evidence, the discretion afforded the expert agency should be sustained.

In addition, although not directly applicable to nitrate and nitrite, the State Board has previously held that the Regional Board may use the State Implementation Policy (SIP) as guidance for determining whether and to what extent to allow dilution credits and a mixing zone. (SWRCB Order WQ 2012-0013, at 16.) The SIP, which establishes state policy for permitting discharges of toxic pollutants to surface waters, provides that a Regional Board may deny or limit a mixing zone where necessary “to protect beneficial uses, meet the conditions of this Policy, or comply with other regulatory requirements.” SIP 1.4.2.2(B). Moreover, the SIP is emphatic that “a mixing zone shall not compromise the integrity of the entire water body.” SIP 1.4.2.2(A)(1) (emphasis in original).³

Here, even beyond the additional technical support provided with these comments, it is clear that the Regional Board has weighed the evidence in the record before it and properly proposed to deny the request for a __ mile long mixing zone. In the Tentative Permit, the Regional Board describes how the discharge of nitrate and nitrite may impact municipal and aquatic life beneficial uses. (Permit, at F-22-23, F-56-58.) The Fact Sheet describes several impacts to beneficial uses that can and are occurring due to the discharge of nutrients and concludes that “any increased nutrient loading contributes to the impairment of the beneficial uses” and therefore denies the requested mixing zone for nitrate plus nitrite. In addition, many of the reasons the Regional Board cited for denying the mixing zone are the precise bases for denying a mixing zone outlined in the SIP: adverse impacts to biologically sensitive or critical habitats; objectionable color, odor, taste, or turbidity; undesirable or nuisance aquatic life; and impacts to drinking water intakes. (Permit, at F-23.) The Regional Board concluded that the discharge of nutrients had the reasonable potential to cause or contribute to the Basin Plans’ narrative objectives for biostimulatory substances and taste and odors. (Permit, at F-57.) The Regional Board also found that there is no assimilative capacity for additional loading of nutrients. (*Id.*) This sound analysis should be maintained in the final permit denying the requested mixing zone.

³ According to the SIP, a mixing zone shall not: (1) compromise the integrity of the entire water body; (2) cause *acutely toxic conditions to aquatic life passing through the mixing zone; (3) restrict the passage of aquatic life; (4) adversely impact biologically sensitive or critical habitats, including, but not limited to, habitat of species listed under federal or State endangered species laws; (5) produce undesirable or nuisance aquatic life; (6) result in floating debris, oil, or scum; (7) produce objectionable color, odor, taste, or turbidity; (8) cause *objectionable bottom deposits; (9) cause nuisance; (10) dominate the receiving water body or overlap a mixing zone from different outfalls; or (11) be allowed at or near any drinking water intake.” (SIP at p. 17.)

References

- Archibald Consulting, Palencia Consulting Engineers, Starr Consulting. 2012. California State Water Project Watershed Sanitary Survey, 2011 Update. Prepared for the State Water Project Contractors Authority and the California Department of Water Resources.
- Collos, Y., Siddiqi, M. Y., Wang, A. D., Glass, M., Harrison, P.J., 1992. Nitrate uptake kinetics by two marine diatoms using the radioactive tracer ^{13}N . *J. Exp. Mar. Biol. Ecol.*, 163, 251–260.
- Collos, Y., Vaquer, A., Bibent, B., Slawyk, G., Garcia, N. Souchu, P. 1997. Variability in nitrate uptake kinetics of phytoplankton communities in a Mediterranean coastal lagoon. *Est. Coastal Shelf Sci.*, 44, 369–375.
- Cornwell, J.C., P.M. Glibert, and M. Owens. 2014. Nutrient fluxes from sediments in the San Francisco Bay Delta. *Estuaries and Coasts*. Doi:10.1007/s12237-013-9755-4.
- Department of Water Resources. 2013. The Department of Water Resources Aquatic Pesticides Application Plan, Water Quality Order No. 2013-0002-DWQ.
- De Troch, M., Vergaerde, I., Cnudde, C., Vanormelinger, P., Vyverman, W., Vincx, M., 2012. The taste of diatoms: the role of diatom growth phase characteristics and associated bacteria for benthic copepod grazing. *Aq. Microb. Ecol.* 67: 47-58.
- Downing, J. A., Watson, S. B., McCauley E. 2001. Predicting cyanobacterial dominance in lakes. *Can. J. Fish. Aquat. Sci.*, 58, 1905-1908.
- Flynn K, Franco, J.M., Fernández, P., Reguera, B., Zepata, M., Wood, G., Flynn, K.J., 1994. Changes in toxin content, biomass and pigments of the dinoflagellate *Alexandrium minutum* during nitrogen refeeding and growth into nitrogen and phosphorus stress. *Mar. Ecol. Prog. Ser.* 111, 99-109.
- Glibert, P.M., D. Fullerton, J.M. Burkholder, J.C. Cornwell, and T.M. Kana. 2011. Ecological stoichiometry, biogeochemical cycling, invasive species, and aquatic food webs: San Francisco Estuary and comparative systems. *Reviews in Fisheries Science*, 19(4): 358-417.
- Glibert, P.M., T. M. Kana, and K. Brown. 2013. From limitation to excess: consequences of substrate excess and stoichiometry for phytoplankton physiology, trophodynamics and biogeochemistry, and implications for modeling. *J. Mar. Systems*. 125: 14-28.
- Granéli E, Flynn K., 2006. Chemical and physical factors influencing toxin content. In: Granéli E, Turner, J.T. (Eds.), *Ecology of Harmful Algae*. Springer, Heidelberg, pp. 229-241.
- Ha, J. H., T. Hidaka, and H. Tsuno. Quantification of toxic *Microcystis* and evaluation of its dominance ratio in blooms using real-time PCR. *Envir. Sci. Technol.*, 43: 812-818 (2009).
- Jeyasingh, P. D., and L. J. Weider. Phosphorus availability mediates plasticity in life-history traits and predator-prey interactions in *Daphnia*. *Ecol. Lett.*, 8: 1021-1028 (2005).
- Jeyasingh, P. D., and L. J. Weider. Fundamental links between genes and elements: evolutionary implications of ecological stoichiometry. *Mol. Ecol.*, 16: 4649-4661 (2007).
- Kana, T.M., Geider, R.J., Critchley, C., 1997. Photosynthetic pigment regulation in microalgae by multiple environmental factors: a dynamic balance hypothesis. *New Phytol.* 137, 629-638.

- Lee, S.J., M.-H. Jang, H.-S. Kim, B.-D. Yoon, and H.-M. Oh. Variation on microcystin content of *Microcystis aeruginosa* relative to medium N:P ratio and growth stage. *J. Appl. Microbiol.* **89**: 323-329 (2000).
- Lehman, E. Seasonal occurrence and toxicity of *Microcystis* in impoundments of the Huron River, Michigan, USA. *Water Res.*, **41**: 795-802 (2007).
- Lomas, M. W., Glibert, P. M., 1999a. Interactions between NH_4^+ and NO_3^- uptake and assimilation: comparison of diatoms and dinoflagellates at several growth temperatures. *Mar. Biol.*, **133**, 541-551.
- Lomas, M. W., Glibert, P. M., 1999b. Temperature regulation of nitrate uptake: A novel hypotheses about nitrate uptake and reduction in cool-water diatoms. *Limnol. Oceanogr.*, **44**, 556-572.
- MacIntyre, H. L., Lomas, M. W., Cornwell, J., Suggett, D. J., Gobler, C. J., Koch, E. W., Kana, T. M., 2004. Mediation of benthic-pelagic coupling by microphytobenthos: An energy-and material-based model for initiation of blooms of *Aureococcus anophagefferens*. *Harmful Algae*, **3**, 403-437.
- McCarthy, J.J., W.R. Taylor and J.L. Taft. 1977. Nitrogenous nutrition of the plankton in the Chesapeake Bay. 1. Nutrient availability and phytoplankton preferences. *Limnol. Oceanogr.* **22**: 996-1011.
- Parker, A.E., W.J. Kimmerer and U.U. Lidström. 2012. Reevaluating the Generality of an Empirical Model for Light-Limited Primary Production in the San Francisco Estuary. *Estuaries and Coasts*, **35**:930-942.
- Sterner, R.W. and J.J. Elser. 2002. Ecological stoichiometry: The biology of elements from molecules to the biosphere. Princeton University Press, Princeton, N.J.
- Tang, K.W. , Taal, M., 2005. Trophic modification of food quality by heterotrophic protists: species-specific effects on copepod egg production and egg hatching. *J. Exp. Mar. Biol. Ecol.*, **318**, 85-98.
- Van de Waal, D. B., J. M. Verspagen, M. Lurling, E. Van Donk, P. M. Visser, J. Huisman. The ecological stoichiometry of toxins produced by harmful cyanobacteria: An experimental test of the carbon-nutrient balance hypothesis. *Ecol. Lett.*, **12**: 1326-1335 (2009).
- Vézic, C., J. Rapala, J. Vaitomaa, J. Seitsonen, and K. Sivonen. Effect of nitrogen and phosphorus on growth of toxic and nontoxic *Microcystis* strains and on intracellular microcystin concentrations. *Microb. Ecol.* **43**: 443-454 (2002).

Technical Memorandum Nutrient Science Summary

February 1, 2014

Historically, scientists have described primary productivity in the Bay-Delta as not being limited by nutrients (Cloern 2001; Lopez et al. 2006) and not experiencing signs of classic eutrophication (Cloern 2001). Such classic eutrophication effects include increased chlorophyll *a* (chl-*a*) in the water column (i.e., more algal blooms), development of hypoxia or anoxia (low or no dissolved oxygen, respectively), loss of native submerged aquatic vegetation, increased proportion of those algae that are considered harmful algal blooms (HABs), and changes in biodiversity, including loss of certain fisheries (e.g., Cloern 2001; Anderson et al. 2002). However, we now recognize that changes in nutrient loads are affecting ecosystem dynamics in complex ways that extend beyond our historic understanding of the process of eutrophication.

Nutrient effects on aquatic systems are far more complex than those normally associated with eutrophication and can occur across the full spectrum of nutrient limitation to nutrient super-saturation (Glibert et al 2013). Even relatively small changes in nutrient supply – even when those nutrients are not limiting for primary production – are being shown to have large consequences on many important properties of ecosystems (Nielsen, 2003). Two important properties of nutrient supply that have effects beginning at the level of the physiology of primary producers (algae) and propagating through the food web are the form in which nutrients are supplied (chemical state, oxidized vs. reduced, organic vs. inorganic, dissolved vs. particulate) and the proportion of different elements (including carbon (C), nitrogen (N), phosphorous (P), and silicon (Si), among others). In the Bay-Delta, the total loads, the forms, and the relative proportions of nutrients have been changing over time. These changes have had profound effects on ecosystem structure of this system, as documented below. This technical memorandum describes the effects of two such changes: the proportion of ammonium to nitrate ($\text{NH}_4\text{:NO}_3$) and the proportion of nitrogen to phosphorus (N:P).

Ammonium:Nitrate

For decades, researchers have explored the relative use – or relative preference for – different forms of nitrogen (N) by phytoplankton. Ammonium (NH_4) is generally considered to be the form of nitrogen preferred by phytoplankton due to the more favorable energetics associated with its assimilation compared to that of nitrate (NO_3). Ecological consequences of dependence on NO_3 vs. NH_4 have also been well recognized for decades. Based on the concept of “new” and “regenerated” production (Dugdale and Goering 1967, Eppley and Peterson 1979), phytoplankton dependence on NH_4^+ leads to production that is cycled within the microbial loop, whereas that based on NO_3^- more often leads to production that supports a food web leading to secondary production as well as export out of the euphotic zone (Dugdale and Goering, 1967; Eppley and Peterson, 1979). Shifts in nitrogen (N) form from NO_3^- to NH_4^+ have been shown in numerous systems to lead to community shifts away from plankton communities dominated by diatoms to those dominated by flagellates, cyanobacteria, and bacteria, in turn, resulting in a shift in composition of higher food webs (e.g., Legendre and Rassoulzadegan, 1995; Glibert, 1998; Mousseau, 2001; Heil et al., 2007). Until recently, most of the data illustrating such changes resulting from shifts in nutrient form were derived from systems for which N was the limiting nutrient. An important question in this context is whether the physiological and ecological consequences of dependence on NO_3^- versus NH_4^+ remain the same under nutrient rich conditions as under conditions of nutrient deficiency. A classic assumption is that when cells are growing at maximal growth rates (set by environmental conditions of light, temperature, etc.), it is fully expected that the total N taken up by cells will be the same whether they are provided NO_3^- or NH_4^+ (or urea or other forms of N). In some nutrient rich (but not classically

eutrophic) estuaries receiving high loads of NH_4 there is evidence for reduced, rather than increased, rates of primary productivity, compared to systems receiving comparable N loads in oxidized forms; such systems have been termed High Nutrient Low Growth (HNLG) systems (Yoshiyama and Sharp 2006).

Although nutrient effects have generally not been considered controlling factors in San Francisco Estuary, the more subtle ecological impacts of NH_4^+ loading and the importance of changes in $\text{NO}_3^-:\text{NH}_4^+$ in phytoplankton succession are beginning to be considered as important factors that may have contributed to historical changes seen in the food web (e.g., Dugdale et al., 2007; 2012, 2013; Glibert, 2010; 2012; Glibert et al., 2011; Parker et al., 2012a,b). Not only have dominant species changed in this system, but rates of primary production have also declined over the course of the past few decades (e.g., Jassby et al., 2002; Kimmerer et al., 2012). Without question, the Bay Delta is receiving high loads of NH_4 and these loads have increased over the recent decades (Fig. 1). A question receiving considerable attention is whether these concentrations of NH_4 have contributed to the tilting of the Bay-Delta away from a productive ecosystem to one that has HNLG characteristics. Based solely on previously reported ambient chl *a* in Suisun Bay values that tend to be $<5 \mu\text{g L}^{-1}$ on average (occasional blooms excepted), combined with ambient nutrient concentrations, this system is characteristic of an HNLG region (Yoshiyama and Sharp, 2006; Dugdale et al., 2007).

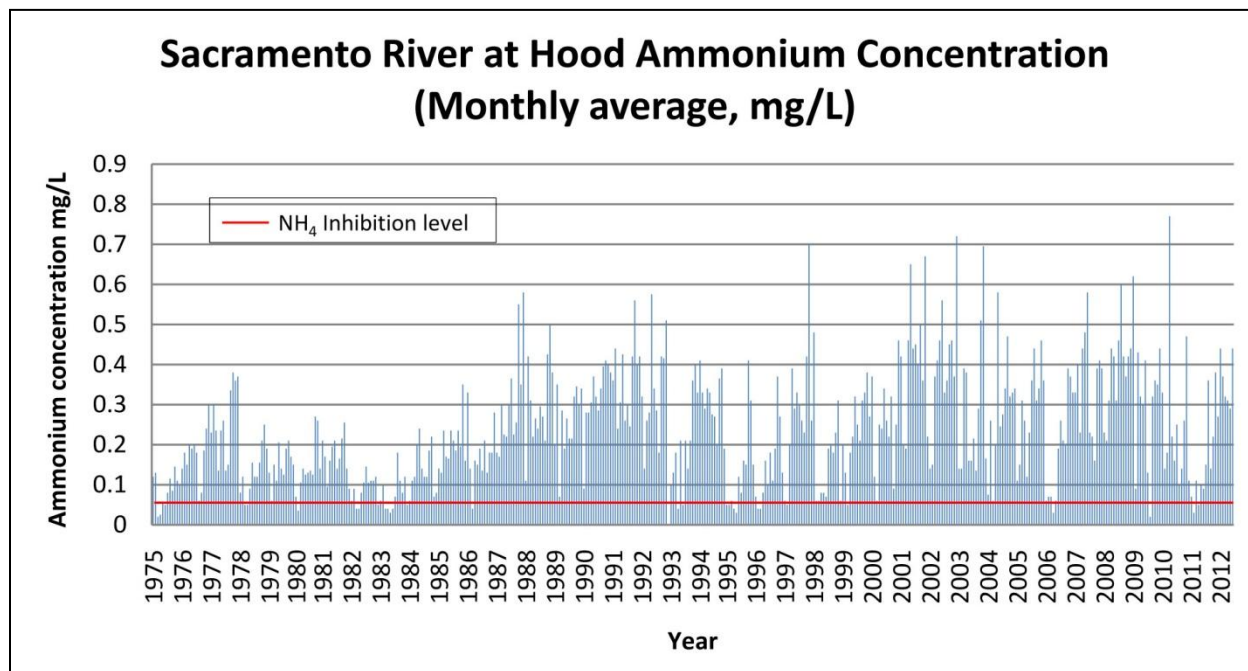


Figure 1. Average monthly ammonium concentration in the lower Sacramento River at Hood. Source: Environmental Monitoring Program data.

The fundamental mechanism contributing to HNLG systems is the inhibition of NO_3^- uptake by NH_4 . More correctly termed “repression” rather than “inhibition”, this phenomenon is well documented throughout the physiological literature. Both NH_4^+ and NO_3^- are transported across the cell membrane by both passive and active pathways. Active pathways involve transporters, specific proteins that transport the targeted molecules. For most algal functional groups, once transported into the cell, NO_3^- is first reduced to NO_2^- through the activity of Nitrate Reductase (*NR*) and subsequently further reduced to NH_4 , the form that can be assimilated into amino acids and proteins for cell growth. Both the process of NO_3^- transport into the cell and its subsequent reduction to NH_4 can be inhibited (or repressed) by NH_4 . Because of differences in the physiology of transporters and cell metabolism between different classes of

algae, different types of algae have different susceptibilities to NH_4^+ . Such differences contribute to the confusion about, different observations of, and different interpretations as to the importance of, NH_4 in regulating productivity of algae.

The inhibition or repression of NO_3^- uptake by NH_4^+ needs to be differentiated from the metabolic effect of toxicity of NH_4^+ or NH_3 . *NH_4^+ is, in fact, best characterized as a paradoxical nutrient – preferentially used at one end of the concentration spectrum when N is limiting and toxic to the cell when supplied at super-saturating levels* (Britto and Kronzucker 2002). Whereas NH_4^+ is transported across the cell membrane via active transport, the unionized form, NH_3 , can diffuse through membranes (Kleiner 1981). The presence of NH_3 increases with increased pH, a condition that is not the norm in aquatic systems, except under dense blooms. More about toxicity effects of NH_4 and NH_3 can be found in the Total Ammonia Toxicity section below.

NH_4 suppression of NO_3 uptake when both nutrients are in ample supply must also be differentiated from the preferential use of NH_4 by phytoplankton when N is limiting. Under the latter conditions, phytoplankton will use NH_4 preferentially because it requires less energy than NO_3 . Under the former conditions, the cells must cope with an excess; and in doing so, their metabolism is less capable of assimilating NO_3 .

The effects of changes in the proportion of $\text{NH}_4:\text{NO}_3$ have been shown for the Bay-Delta in both field observations and laboratory experiments. Parker et al. (2012a) observed a 60% decline in primary production in the Sacramento River below the Sacramento Regional Wastewater Treatment Plant, where NH_4 is discharged, compared to production above the Treatment Plant's outfall. Also supporting this finding, Parker et al. (2012b) found that “[b]y tracing both carbon (C) and N uptake we provide clear evidence that high rates of C uptake are linked to phytoplankton NO_3 , and not NH_4 , use.” They conclude that the increased proportion of NH_4 “may help explain some of the reduced primary production and phytoplankton biomass observed [in the San Francisco Estuary] since the 1970s.”

In enclosure experiments with samples from Central Bay, Suisun Bay, and the Sacramento River at Rio Vista, representing a gradient of both nutrient concentrations and proportions of different forms of N, Wilkerson et al. (in preparation) observed “a gradient of decreasing phytoplankton physiological rates in the upstream direction as far as Rio Vista.” Phytoplankton productivity rates (both carbon and nitrogen uptake) decreased with increasing concentrations of NH_4 .

In a series of recent experiments conducted with natural samples from the Sacramento River, the effects of altered proportions of $\text{NH}_4:\text{NO}_3$ were also apparent (Glibert et al. 2012b). In these experiments, the proportions of these nutrients were manipulated, and both short-term N uptake rates and longer-term N production rates were assessed. Two findings are of note. First, when 20 μmol NH_4 was added to the sample, a concentration commonly observed in the Sacramento River, and the rate of NO_3 uptake was measured across a concentration gradient, the rate of uptake of NO_3 decreased significantly compared to unamended rates measured over a period of < 1 hour (Figure 2). Second, when samples were enriched with NH_4 , NO_3 , or urea (at the molar equivalent dose) for a period of 24-48 hours, and then rates of uptake of all N forms measured, the summed rate of N uptake in the NH_4 -added treatment was significantly lower than that in the NO_3 -added or urea-added treatments (Figure 3).

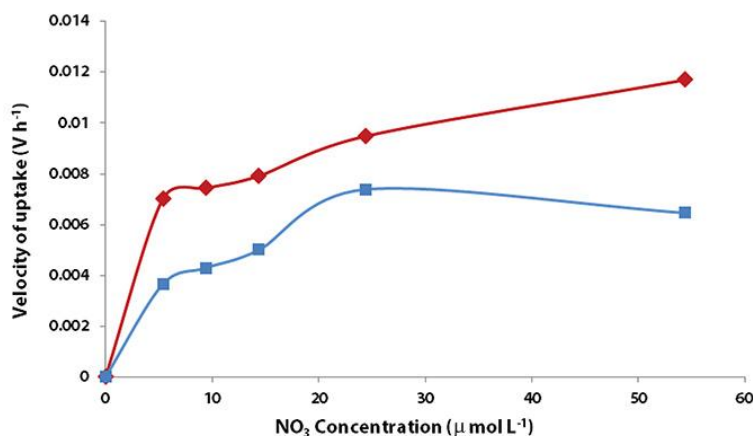


Figure 2. Velocity of uptake of NO₃ as a function of added NO₃ enrichment (red curve), and the same relationship but with a constant addition of 20 μmol L⁻¹ NH₄ (blue curve). Nitrate uptake is reduced when NH₄ is added. Experiment was conducted with water collected from the Sacramento River. Data from Glibert et al. (2012b).

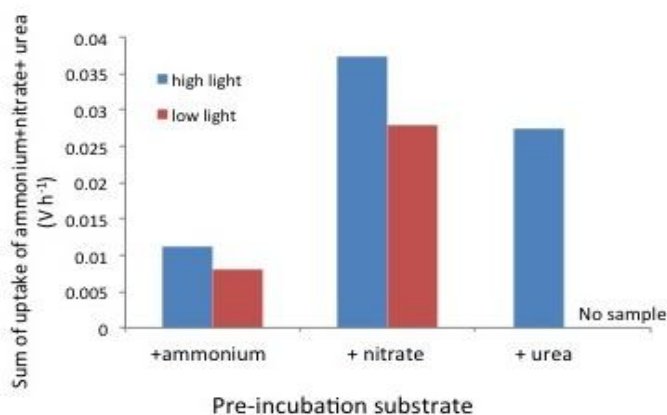


Figure 3. Summed rate of uptake of nitrate+ ammonium+ urea for samples collected from the Sacramento River and pre-incubated with the substrate indicated, after which short-term uptake rates were measured using ¹⁵N tracer techniques. The experiment was conducted under both high (blue bars) and low (red bars) irradiance levels. The summed rate of N uptake in the NH₄-added treatment was significantly lower than that in the NO₃-added or urea-added treatments. From Glibert et al. (2012b).

Longer term responses were also found in paired experiments in which NO₃ and NH₄ concentrations were manipulated for San Francisco Bay-Delta samples collected over 2 years, leading to different types of phytoplankton developing over a period of several days of exposure. A greater response by fucoxanthin-containing organisms (diatoms) was observed in those samples enriched with NO₃⁻, and greater responses by zeaxanthin and Chl *b*-containing organisms (predominantly cyanobacteria and chlorophytes) were observed in samples enriched with NH₄⁺ (Glibert et al., in review). A series of 3-week nutrient-rich mesocosm experiments conducted in Wascana Lake, Saskatchewan, Canada, yielded largely similar findings: total cyanobacterial biomass was associated with NH₄⁺ treatments and diatom biomass was associated with NO₃⁻ treatments (Donald et al., 2013). In recent experiments conducted in the tidal freshwater estuarine zone of the Guadiana Estuary (Spain and Portugal), it was also found that NO₃ consumption decreased with increasing NH₄ uptake, and these findings were most pronounced during the

most productive period. Total primary productivity was suppressed as a result (Domingues et al. 2011). As stated by Domingues et al. (2011), "...increased inputs of N as NH_4 due to urban waste effluents may result in a shift in phytoplankton community composition, towards a dominance of cyanobacteria and green algae."

This finding is further supported by the 37 years of data collected by the Environmental Monitoring Program in the Bay-Delta. When monthly data of chl-a and diatom cell count are plotted against NH_4 levels for the period 1975 to 2012 there is a marked decreasing trend in both as ammonium levels rise, with an increasing effect around the 0.056 mg L^{-1} ($4 \mu\text{mol L}^{-1}$) level (Figures 4 and 5). This level of NH_4 has been identified as a threshold value above which inhibition or repression of NO_3 uptake begins (Dugdale et al. 2007).

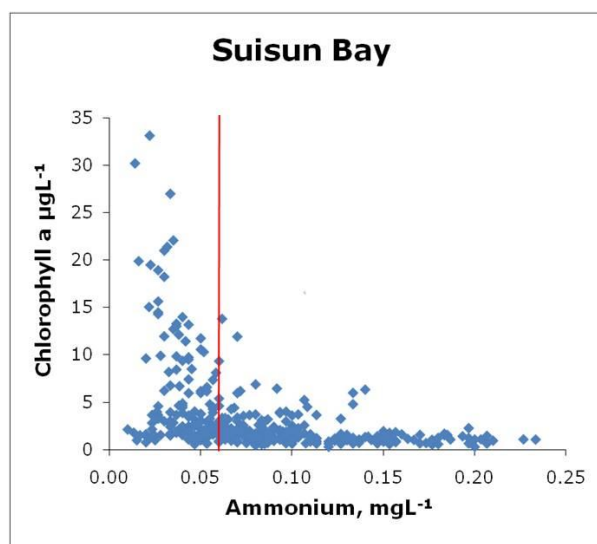


Figure 4. Chlorophyll-*a* concentration plotted with ammonium concentration in Suisun Bay. As NH_4 concentration increases above the level where inhibition is observed (red line at 0.056 mg L^{-1} NH_4), chl-*a* levels decline. (Source: Environmental Monitoring Program data)

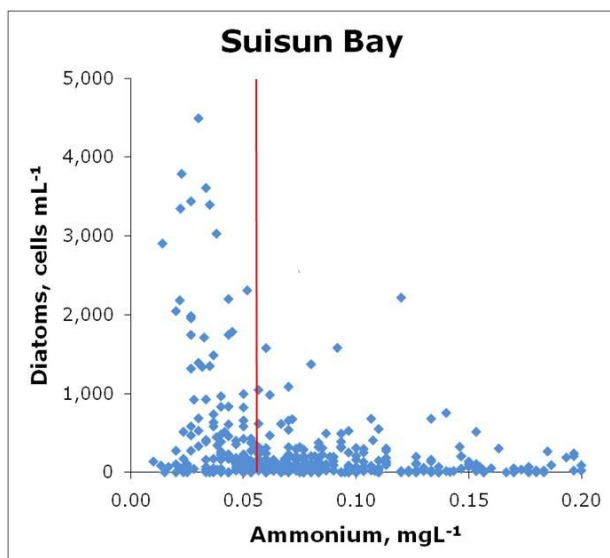


Figure 5. Diatom cell count plotted with ammonium concentration in Suisun Bay. As NH_4 concentration increases above the level where inhibition is observed (red line at 0.056 mg L^{-1} NH_4), diatom abundance declines. (Source: Environmental Monitoring Program data)

Similar threshold values have been reported by others. Lomas and Glibert (1999) described the threshold for inhibition of NO_3^- uptake at NH_4^+ levels of approximately $1 \mu\text{mol L}^{-1}$ (0.014 mg L^{-1}). Yoshiyama and Sharp (2006) saw a “striking decline in production at NH_4^+ levels above a low threshold (around $10 \mu\text{mol L}^{-1}$)” (0.14 mg L^{-1}).

The Delta’s algal species composition has shifted over the past decades from diatoms to smaller and lower quality food species such as flagellates, cryptophytes and cyanobacteria (Lehman, 2000; Lehman *et al.*, 2005; Lehman *et al.*, 2010; Jassby *et al.*, 2002; Sommer *et al.*, 2007; Glibert, 2010; Glibert *et al.*, 2011; Winder and Jassby, 2010). Dugdale *et al.* (2012) developed a conceptual model that correctly predicted the development of two rare, spring phytoplankton blooms in Suisun Bay based on only three criteria: the rate of NH_4^+ loading (based on present day sewage effluent loads), the water column concentration of NH_4^+ , and river flow (analogous in steady-state chemostat growth to “washout”). This conceptual model was further advanced in a one-dimensional, N-based model (Dugdale *et al.*, 2013), having the unique features of the inclusion of terms for the time-varying rates of maximum NO_3^- uptake as a function of NO_3^- concentration and for inhibition of NO_3^- uptake by NH_4^+ . The modeled high-biomass, NO_3^- -based, high-productivity state is analogous to the pre-1982, diatom era of the Bay-Delta. The importance of NH_4^+ inhibition of NO_3^- uptake was considered to be a necessary interaction to include in a recent model of the emergent phytoplankton community in the California Current System (Goebel *et al.* 2010).

The evidence is continuing to mount that the increasing loads of NH_4^+ are an important contributor to the observed changes in phytoplankton community. The emerging experimental data – assembled from samples collected from the Bay-Delta and elsewhere- provide direct experimental evidence that dichotomous phytoplankton communities develop when enriched with the same absolute concentration of NO_3^- and NH_4^+ , even when sufficient N nutrient was available to the community prior to the N inoculations. Although there is much yet to be learned about the role of nutrient control in many aspects of the Bay-Delta ecosystem, the patterns of response by algal communities to NH_4^+ vs. NO_3^- are consistent with observations in other systems and are consistent with the differential physiology of different algal groups.

Total Ammonia Toxicity

In addition to altering phytoplankton community structure, growth rates and abundance, ammonia is also toxic to some higher trophic level organisms. Scientists at University of California, Davis have investigated the effects of total ammonia nitrogen ($\text{NH}_3 + \text{NH}_4^+$) on the growth, reproduction and survival of the calanoid copepod *Pseudodiaptomus forbesi* using a full life-cycle bioassay approach (Teh *et al.* 2011). *P. forbesi* is an important prey item for the young of many fish species in the Bay-Delta including delta smelt and longfin smelt (Nobriga 2002; Hobbs *et al.* 2006; Feyrer *et al.* 2003). Teh *et al.* (2011) found that total $\text{NH}_3 + \text{NH}_4^+$ at 0.36 mg L^{-1} ($25.7 \mu\text{mol L}^{-1}$) significantly affects the recruitment of new adult copepods, and that total $\text{NH}_3 + \text{NH}_4^+$ at 0.38 mg L^{-1} ($27.1 \mu\text{mol L}^{-1}$) significantly affects the number of newborn nauplii surviving to 3 days (Teh *et al.* 2011). For comparison, monthly water samples collected between 2009-2010 from the Sacramento River between Hood and Isleton, approximately 30 miles downstream from the discharge point for the Sacramento Regional Wastewater Treatment Plant, exceeded the level of $0.36 \text{ mg/L NH}_4\text{-N}$ 44% of the time (Figure 6).

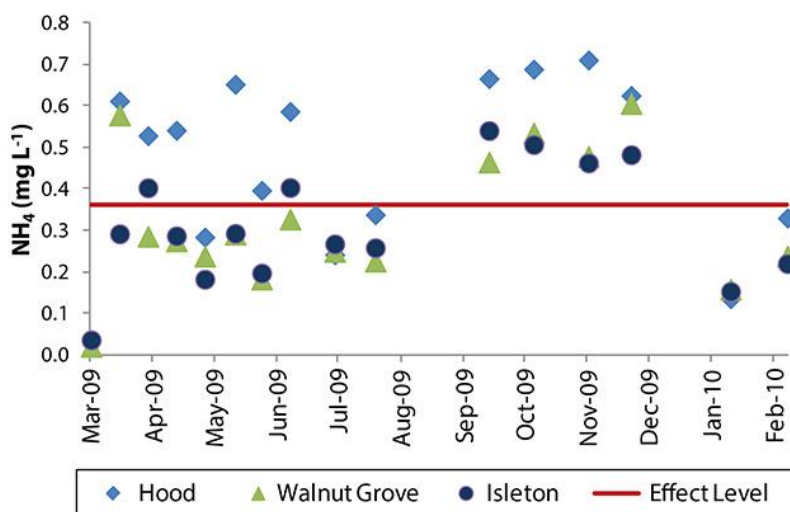


Figure 6. Ammonium concentrations in the Sacramento River measured at Hood, Walnut Grove, and Isleton between 2009 and 2010. Hood and Isleton are approximately 8 and 30 miles downstream of the discharge, respectively. The horizontal line at 0.36 mg L^{-1} is the level at which significant toxicity to copepods is observed. Data from Foe et al. (2010).

The Teh et al. (2011) study was recently cited in the U.S. Fish and Wildlife Service 12-month finding on the petition to list the Bay-Delta longfin smelt as threatened or endangered under the ESA. (77 Fed. Reg. 19755, 19776 [April 12, 2012].) The proposed rule states that “[a]quatic insects on which the longfin smelt relies upon for food have been shown to be sensitive to ammonia.” (77 Fed. Reg. 19776.) The proposed rule states that “[a]mmonia also can be toxic to several species of copepods important to larval and juvenile fishes.” (*Id.*) The toxic effect of total ammonia is a major stressor on aquatic life that has a pervasive impact across the Bay-Delta estuary.

Further, the U.S. Environmental Protection Agency’s (USEPA) Bay-Delta Action Plan highlights concerns with aquatic life toxicity caused by total ammonia nitrogen and identifies total ammonia levels as one of the suspected contributors to the pelagic organism decline in the Bay-Delta (USEPA 2012). In the Bay-Delta Action Plan, USEPA states that they will finalize the new national ammonia aquatic life criteria and they encourage the Regional Boards to consider adopting the criteria and using the criteria to develop effluent limitations for ammonia. USEPA adopted the new aquatic life water quality criteria for ammonia in 2013, which are more stringent than previous criteria adopted in 1999 and consider ammonia toxicity to freshwater mussels (USEPA 2013).

Nitrogen:Phosphorus

Extensive research has found that the N:P ratio also has profound effects on community structure. The principles of ecological stoichiometry suggests that different organisms will dominate under different relative proportions of critical elements (C, N, or P) due to differences in allocation of C, N and P in the various structures that form the biomass of different types of organisms (Sterner and Elser, 2002). As noted by Hall (2009, p. 504), “Ecological stoichiometry formalizes what should be obvious: Organisms interacting in food webs are composed of different elements, such as C, N, or P. As a result, energy and nutrient flow through consumer-resource interactions obey fundamental constraints.” Elemental differences in biomass are found at all levels of organismal structure across trophic levels, from the subcellular to the macrocellular structural components (Sterner and Elser, 2002). The Redfield ratio

(Redfield, 1934, 1958), in which organismal C:N:P ratios are assumed to be in the proportion of 106:16:1 by atoms, is likely the most well-known stoichiometric relationship.

The N:P ratio of nutrients has doubled in the Bay-Delta estuary over the last 35 years as is apparent from the data on Figure 7. These increases are a result not only of the increasing total N load (due to increasing effluent as well as other sources), but also as a function of declining P loads (Van Nieuwenhuysen 2007; Glibert 2010, Glibert et al. 2011).

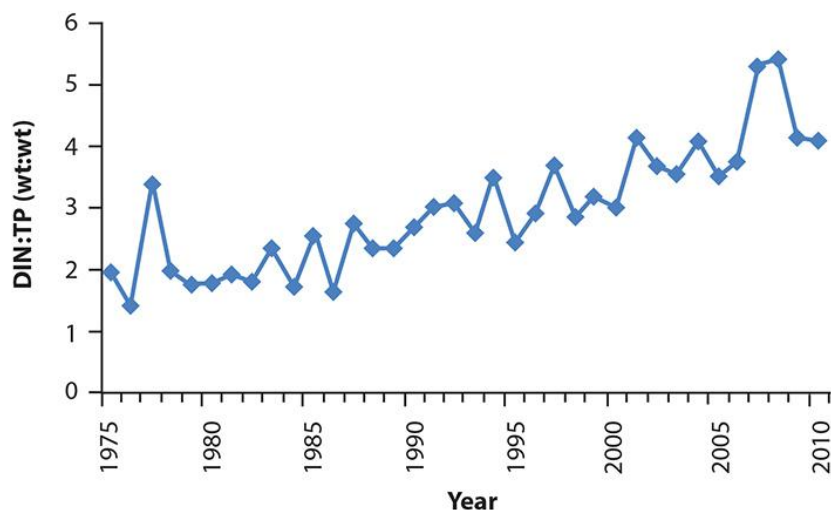


Figure 7. Average annual (March-November) ratio of dissolved inorganic nitrogen to total phosphorus has doubled in the Bay-Delta. (Source: IEP monitoring data from stations in Suisun Bay and the confluence (D4, D6, D7, D8)).

There are a number of strategies available to different types of phytoplankton for coping with an environment where nutrient ratios are not in proportion to their internal requirements (Glibert and Burkholder 2011). For example, cell size is an important determinant of elemental composition (Harris 1986; Finkel et al. 2010). Small cells have a lower requirement for P due to the smaller need for structural components in the cell (Finkel et al. 2010). In comparison to diatoms, very small cyanobacteria such as *Synechococcus* have a much larger cellular ratio of carbon to phosphorus (C:P), on average (Finkel et al. 2010). This explains why small cells, such as *Synechococcus*, have been found to thrive in waters that are comparatively P poor, as is the case in Florida Bay (Glibert et al. 2004).

These basic principles of algal response to changing N:P have relevance to San Francisco Bay-Delta phytoplankton communities. Jassby (2008) stated:

A decrease in percentage of diatom biovolume occurred during 1975–1989, caused by both a decrease in diatoms and an increase in green algae, cyanobacteria, and flagellate species biovolume (Kimmerer 2005; Lehman 1996), i.e., probably in the direction of declining nutritional value per unit biomass. In principle, the total nutritional value of a community could decrease even as its biomass increases. Moreover, changes in size, shape, and motility of species comprising the phytoplankton community could also affect their availability as food particles for crustacean zooplankton and other consumers.

There is strong support in the scientific literature for the proposition that the N:P ratio influences phytoplankton community composition. For example, in the Seto Inland Sea of Japan, removal of

phosphorus also led to a shift in phytoplankton community structure from “nonharmful diatoms to harmful raphidophytes...and then finally to harmful/toxic dinoflagellates” (Yamamoto 2002). In this case the reduction in phosphorus which increased N:P, led to a change in phytoplankton community composition and was suggested to be “the major cause of the reduction in fishery production” (Yamamoto 2002).

In a retrospective analysis of 30 years of data from the Bay-Delta estuary, Glibert et al. (2011) found that the variation in these nutrient concentrations and ratios is highly correlated to variations in the total amount and composition of phytoplankton. This analysis revealed relationships between biological parameters and nutrients and/or nutrient ratios using both the original data and data that were adjusted for autocorrelation. At the phytoplankton level, as described earlier, there has been a decline in total chl-a and a decline in total diatoms over the past several decades in proportion to the increase in total inorganic N to total P (Figure 8). The change in chl-a with N:P is apparent in different regions of the Bay-Delta; as N:P increases, chl-a declines (Figure 9).

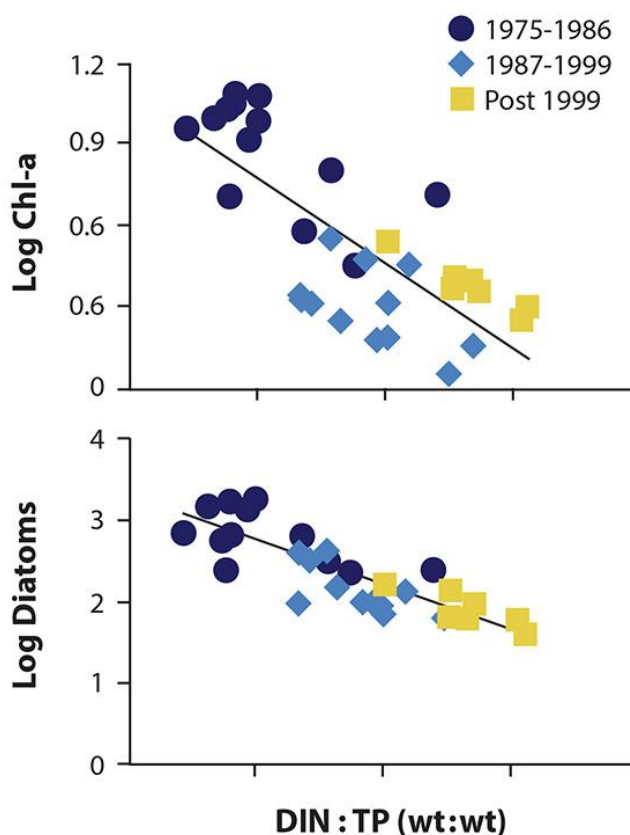


Figure 8. Change in the concentration of chl-*a* ($\mu\text{g L}^{-1}$) and abundance of diatoms (Bacillariophyceae, cells mL^{-1}) as a function of dissolved inorganic N to total phosphorus. A loss of total chl-*a* and a loss of total diatoms in the phytoplankton community have occurred over the past several decades in proportion to the change in total inorganic N to total P. The relationship is significant at $p < 0.05$. Different periods of time are represented by different symbols: 1975-1986, filled circles; 1987-1999, diamonds; post-1999- filled squares. Data shown are for the years 1975-2005 and cover the region from the confluence to Suisun Bay. All data log-transformed. Data from Glibert et al. (2011).

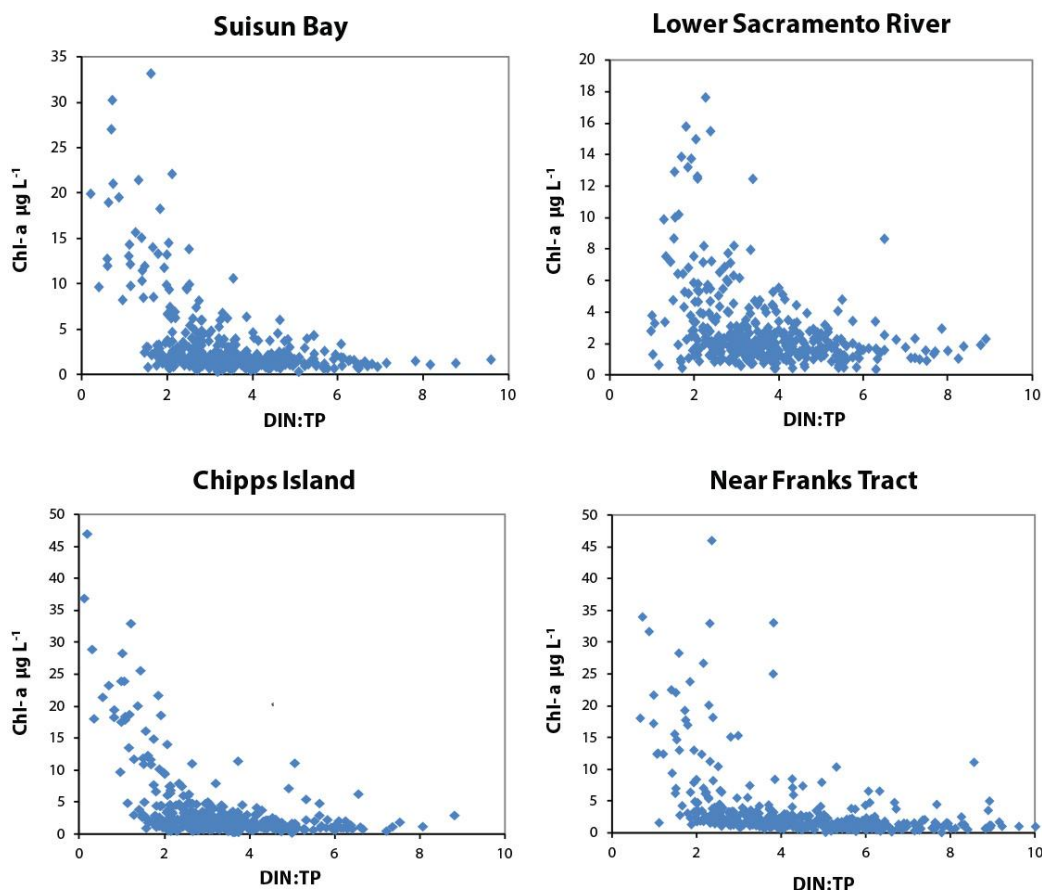


Figure 9. Chl-*a* concentration plotted against DIN:TP for subregions of the Delta, 1975-2011. As DIN:TP increases, there is a loss of chl-*a*. (Source: Environmental Monitoring Program data).

Fast-growing phytoplankton require proportionately more P to satisfy metabolic demands. Diatoms are typically fast-growing, and thus require proportionately more P to meet this metabolic demand. In ecological terms, they are considered a *r*-selected group, would be expected to have a low N:P biomass ratio (due to the high P cellular demand), and thus would be expected to be outcompeted if N:P in the environment increases. So-called *r*-selected species are out-competed when the environment changes (e.g., Heckey and Kilham 1988). In contrast, many cyanobacteria are considered to be *k*-selected, implying a slower growth rate and a higher metabolic N:P. In fact, “Reynolds (1984) singled out *Microcystis* as an example of a *k*-selected phytoplankter because it grows slowly in nature” (Heckey and Kilham 1988).

The balance of N:P can also affect other metabolic aspects of phytoplankton besides growth, including toxin production, cell membrane thickness, and other chemical constituents that have been considered to turn good food “bad” (Mitra and Flynn 2005). At the primary producer level, for example, emergent properties that can change in response to changes in elemental proportions include the relative proportions of ribosomes, enzyme activities, gene regulation, toxin production, cellular pigmentation complement, and ultimately the cell elemental composition, specifically, chl:carbon (Chl:C), C:N, and N:P (Glibert et al. 2013). Toxin production by numerous harmful algae has been shown to increase when the cells are grown under nutrient-imbalanced conditions and when there is a change in N or P availability (Flynn et al. 1994; Johansson and Granéli 1999; Granéli and Flynn 2006). In Daechung Reservoir, Korea, researchers found that toxicity of cyanobacteria was related not only to an increase in N in the water, but

to the cellular N content as well (Oh et al. 2000). A recent report by Van de Waal et al. (2009) demonstrated in chemostat experiments that under high carbon dioxide and high N conditions, microcystin (an algal toxin) production was enhanced in *Microcystis*. Similar relationships were reported for a field survey of the Hiro-sawa-no-ike fish pond in Kyoto, Japan, where the strongest correlations with microcystin were high concentrations of NO_3 and NH_4 and the seasonal peaks in *Microcystis* blooms were associated with extremely high N:P ratios (Ha et al. 2009). Thus, not only is *Microcystis* abundance enhanced under high N:P, but its toxicity appears to be as well (Oh et al. 2000).

It is well accepted that the nutritional value of phytoplankton differs from one species to another. Toxin production can inhibit grazing. Some phytoplankton species are rejected by grazers due to their size. Others vary in their nutritional quality. For example, some diatom species produce certain highly unsaturated fatty acids that are essential for zooplankton reproduction (reviewed by Kilham et al. 1997) while flagellates generally produce different fatty acids than diatoms (Olsen 1999). Many trophic interactions, such as rates of growth or fecundity, are dependent on the acquisition of particular fatty acids, as a measure of the food quality of algae (e.g., Ahlgren et al. 1990; Coutteau and Sorgeloos 1997; Weers and Gulati 1997; Brett and Müller-Navarra 1997). In feeding experiments, Ger et al. (2010) observed reduced survival of the copepods, *Pseudodiaptomus* and *Eurytemora*, even when *Microcystis* was only a small portion of their available diet. Brett and Müller-Navarra (1997) developed a food quality rank for 10 species from 5 major phytoplankton groups based on the average of the observed change in the abundance of individual zooplankters that preyed upon these phytoplankton in growth bioassays. They and others (see Park et al. 2003) have applied a 0-1 scale of phytoplankton food quality in which cyanobacteria ranks at 0.2; green algae, 0.525; diatoms, 0.7; and cryptomonads, 0.95. Thus, a trend of decreasing diatoms and increasing cyanobacteria in the Bay-Delta would suggest, based on these rankings, a decrease in food quality for higher trophic levels.

Cloern and Dufford (2005) state, “[t]he efficiency of energy transfer from phytoplankton to consumers and ultimate production at upper trophic levels vary with algal species composition: diatom-dominated marine upwelling systems sustain 50 times more fish biomass per unit of phytoplankton biomass than cyanobacteria-dominated lakes.”

For species that prey on phytoplankton (e.g., zooplankton), stoichiometry affects all aspects of behavior, such as growth rate, fecundity, and ultimately the success of different populations (Jeyasingh and Weider 2005, 2007), but may affect various life stages differently (Moe et al. 2005, p.31): “[a]n organism’s requirements for different elements may vary throughout its life cycle, and thus certain life stages may be more sensitive than others to variation in the stoichiometry of its resource.” For example, copepod juveniles have a relatively high demand for C, N, and P, but at a later stage, while C is still needed for metabolism, more P must be allocated to eggs. Therefore, P-poor food sources can disproportionately affect egg production while not affecting survival (Faerovig and Hessen 2003; Laspoumaderes et al. 2010). In a laboratory study where *Acartia tonsa* was fed diatoms grown on different N concentrations, Kiørboe (1989), confirmed that this zooplankter changes its feeding rate in response to phytoplankton of different chemical composition – thus, in response to food quality. Moreover, egg production followed the variation in algal N content and increased with increasing algal N. In the Bay-Delta, Slaughter and Kimmerer (2010) observed lower reproductive rates and lower growth rates of the copepod, *Acartia* sp. in the low salinity zone compared to taxa in other areas of the estuary. Their observation may be due to differences in nutrient stoichiometry between these areas.

In a review of field and laboratory-based research on stoichiometry in food webs, Hessen (1997) showed that a shift from copepods to *Daphnia* tracked N:P; copepods retain proportionately more N, while *Daphnia* are proportionately more P rich. Often, those organisms that are most able to retain the nutrient in limited supply, in this case P, have the competitive advantage in an unbalanced system. Glibert et al. (2011) illustrated a finding similar to Hessen’s, that the decline in calanoid copepods in the Bay-Delta,

and the invasion of cyclopoids tracked N:P over time. Variation in proportional densities of the calanoid copepod *Eurytemora* with the cyclopoid copepod *Limnocalanus* over time has followed changes in the DIN:TP (dissolved inorganic nitrogen to total phosphorus) ratio (Figure 10), a pattern consistent with these grazers being responsive to changes in elemental stoichiometry and maintenance of altered dynamic equilibria on a long-term scale. In fact, Glibert et al. (2011) found relationships between many zooplankton species and nutrient composition. Results from whole-lake experimentation suggest that the N:P ratio is linked to alterations in zooplankton size, composition, and growth rate, as those animals with increased RNA allocation (more P available for growth) will grow at higher rates due to increased protein synthesis rates (Sterner and Elser 2002, Schindler 1974). Similar findings were reported from annual studies in the Baltic Sea (Walve and Larsson 1999).

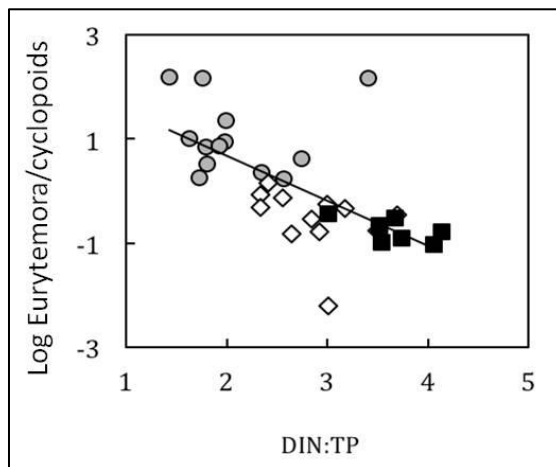


Figure 10. Change in the ratio of *Eurytemora* to cyclopoid copepods (all data log transformed) as a function of DIN:TP for annually averaged data from 1975-2005 for samples collected between the confluence and Suisun Bay. As DIN:TP increases, the proportion of *Eurytemora* to Cyclopoids decreases. Different periods of time are represented by different symbols: 1975-1986, filled circles; 1987-1999, diamonds; post-1999 filled squares. The correlation for these and for data that were detrended (not shown) are significant ($p < 0.05$). From Glibert et al. 2011.

Superimposed on these empirical observations is consideration of whether substrate quality or food quality is altered on an episodic basis, or whether changes are long-term and sustained. As conceptualized by Hood and Sterner (2010), a change in predator growth rate depends on the extent to which a diet is sustained or switches between low-quality food and high-quality food as defined by the relative P content.

Higher Trophic Level Effects

Disproportionate N and P loads are now recognized to have effects at all scales, from genomic to ecosystems that need further empirical resolution (Peñuelas et al. 2012). When N:P availability changes, food webs change, biogeochemical cycling can change, and these changes can be positively reinforcing. Sterner and Elser (2002) state: "[s]toichiometry can either constrain trophic cascades by diminishing the chances of success of key species, or **be a critical aspect of spectacular trophic cascades with large shifts in primary producer species and major shifts in ecosystem nutrient cycling**" [emphasis added]. Just as different elemental ratios may affect the composition of the primary producers, different nutrient requirements of organisms occupying higher trophic levels will have an impact on their ability to thrive as community composition changes at the base of the food web. At the ecosystem scale, the total load and balance of nutrient elements have effects that propagate through the food web, with the potential of transforming ecosystems to new stable states. Although the shift in algal community composition in terms of diatoms and cyanobacteria has been emphasized above, this shift in the Bay-Delta estuary has been far

more complicated. With the decline in water column chl-*a* and an increase in light availability, other primary producers have increased in abundance, including invasive macrophytes such as *Egeria densa* (Sommer et al. 2007; Nobriga et al. 2005; Glibert et al. 2011). *E. densa* may be particularly well suited to the low DIP:DIN environment of the Bay-Delta since it is able to access sediment bound phosphorus through its roots. In fact, similar increases in macrophytes were observed in many other systems in which N:P increased following N enrichment and P reduction, including the Potomac River, Chesapeake Bay, Ebro River in Spain, and the Rhine River in Germany (Glibert et al. 2011; Glibert 2012). Such macrophyte invasions can have profound impacts on ecosystems, not only because they alter the flow of C and the overall productivity of the system, but they also serve as “ecological engineers,” decreasing nutrients through uptake, reducing turbidity by trapping sediments, and providing refuge for zooplankton and habitat for other species, including fish (Yarrow et al. 2009; Glibert 2012).

The interplay between nutrient stoichiometry and biogeochemistry is well illustrated when a system is driven to higher macrophyte productivity. Macrophytes can be highly productive, which can result in elevation of pH due to carbon drawdown in the process of photosynthesis. As noted by Glibert (2012), once pH is elevated, the fundamental physical–chemical relationships related to P adsorption–desorption in sediments change, as does N biogeochemistry (Jordan et al. 2008; Gao et al. 2012). Moreover, under increased pH conditions, the biogeochemistry of calcification is altered, increasing the potential for calcification and the growth of calcifying organisms. Thus, the change in the abundance of the clam *Potamocorbula amurensis* from the time of its introduction in the mid-1980s to 2005 has been shown to be highly and positively correlated to the increase in total N:total P ($r^2 = 0.46$; $n = 20$; $p < 0.01$; all data log transformed), and the average annual abundance of this species has also been found to be highly and positively correlated with mean annual average pH in the estuary ($r^2 = 0.64$; $n = 19$; $p < 0.01$; species abundance data log transformed) (Glibert et al. 2011). Interestingly, the Potomac River, Rhine River and the Ebro River have had similar invasions of macrophytes and *Corbicula* clams that relate to increases in N:P loading (Ibanez et al. 2008; Glibert et al. 2011; Glibert 2012).

In the Bay-Delta estuary, data show top-down grazing of phytoplankton by the clam *P. amurensis* exerts a strong control on phytoplankton biomass, as is also the case for other systems when invaded by bivalve mollusks. Prior interpretations, emphasizing stochastic invasions largely via ballast water exchange imply that the invasive event was the ultimate cause of the change in top-down control of phytoplankton. The ecological stoichiometric interpretation does not preclude strong top-down control of selected component organisms, nor ballast water exchange as the mechanism of introduction. The distinction is that, at the overall ecosystem level, the structuring of species is affected by alterations in nutrients and ecosystem biogeochemistry.

The arguments presented here make the case that bottom-up control contributed to the conditions that allowed *P. amurensis* to become a dominant regulator of phytoplankton production. In other words, invasive species effects and nutrient effects are interrelated. This interpretation is consistent with Ware and Thompson’s (2005) insights from a broad survey of the relative contributions of “bottom-up” vs. “top-down” factors that potentially control fish catch in the coastal waters of the western U.S.; they, too, reported that bottom-up factors were more important.

Several recent reviews have investigated the stoichiometry of fish (Sturner and George 2000; Hendrixson et al. 2007; McIntyre and Flecker 2010). Not only does a strong shift in body N:P occur with growth stage (Pilati and Vanni 2007), but strong differences between taxonomic families also occur. In fact, Hendrixson et al. (2007) demonstrated, for 20 families of fish, that a phylogenetic tree could be developed based on the body nutrient composition.

In the Bay-Delta estuary, numerous changes in fish community composition occurred in relation to phytoplankton and zooplankton changes, and to N:P (Glibert 2010; Glibert et al. 2011) (Figure 11).

Glibert et al. (2011) also found that total P “explained at least as much of the variability in delta smelt as did the [Feyrer et al. 2011] habitat index, and dinoflagellate abundance explained even more.” Unlike correlations with Bay-Delta outflow or with the location of the 2 practical salinity unit isohaline, where the underlying mechanisms driving the correlations are largely unknown, the nutrient relationships have a strong mechanistic explanation in ecological stoichiometry and stable state principles. For this reason, there is relatively low uncertainty that changes in nutrient stoichiometry in the Bay-Delta estuary, achieved through both external forces (altered land-based nutrient loads) and internal, organism-driven, assimilative and dissimilative processes, are related to community compositional changes (Glibert et al. 2011; Glibert 2012).

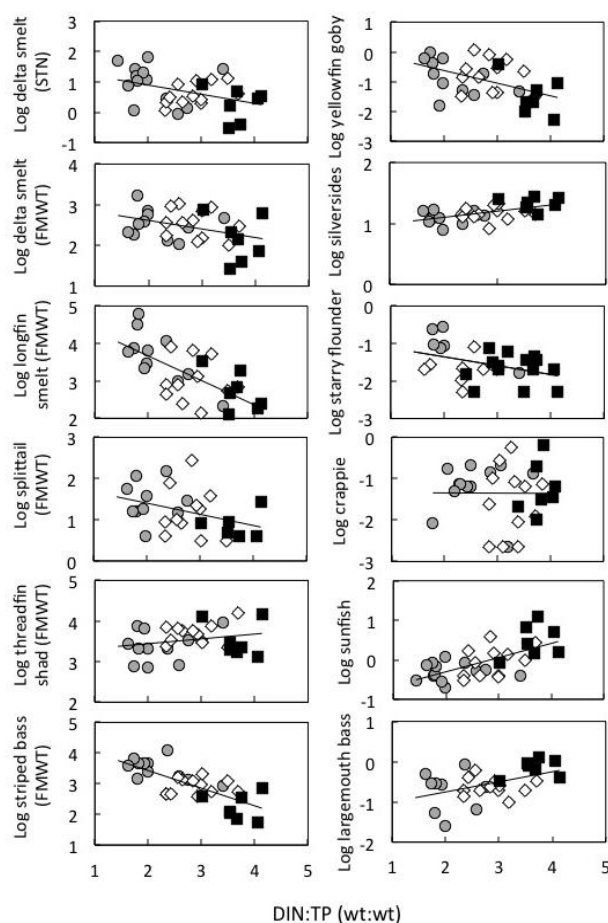


Figure 11. Changes in the abundance of major fishes in relation to ratio of dissolved inorganic nitrogen to total phosphorus from 1975-2005. Different periods of time are represented by different symbols: 1975-1986, filled circles; 1987-1999, diamonds; post-1999 filled squares. All data were log-transformed. The correlations for all fish except crappie were significant ($p < 0.05$) in these data as well as in data that were detrended. Source: Glibert et al. (2011).

Total Nutrient Loads

Total nutrient load sets the upper limit on total primary production, and increases in nutrient loading are commonly associated with eutrophication. Nutrient levels in water diverted from the Delta are at concentrations that can produce nuisance algal and aquatic weed growth and adversely affect drinking water beneficial uses in downstream conveyance facilities and reservoirs. Algal and aquatic plant growth in the SWP conveyance facilities and downstream reservoirs is neither light limited nor inhibited by high

ammonium concentrations since most of the ammonium from the Delta has been nitrified to nitrate. Elevated levels of nutrients (phosphorus and nitrogen compounds) stimulate nuisance algal and aquatic weed growth that includes production, by specific cyanobacteria, of noxious taste and odor compounds and algal toxins. In addition to algal produced taste and odor and algal toxin concerns, increases in algal and aquatic weed biomass can impede flow in conveyances, shorten filter run times and increase solids production at drinking water treatment plants, and add to organic carbon loading.

Frequently annual phosphorus concentrations at Clifton Court Forebay have averaged 0.11 mg/L and total nitrogen has averaged 0.87 mg L⁻¹. Phosphorus is significantly higher than the 0.020 to 0.042 mg L⁻¹ that has been associated with a high risk of nuisance growth and eutrophication (USEPA 1980, 2001a). Levels of both nutrients exceed USEPA Ecoregion I phosphorus and total nitrogen reference conditions of 0.047 mg L⁻¹ and 0.31 mg L⁻¹, respectively (USEPA 2001b)⁴. Ecoregion I includes the Central Valley.

Literature values and USEPA's ecoregion reference conditions⁵ provide a starting point for determining whether nutrient concentrations in Delta waters are at levels that could cause water quality impairments, such as algal production of compounds that produce noxious tastes and odors. More importantly, there is already significant evidence of nutrient-related adverse impacts from Delta water. Through 2006, the Department of Water Resources (DWR) has applied algacide treatments to Clifton Court Forebay for aquatic weeds and algae multiple times each summer. This practice was halted in 2007, however, over concerns of potential impacts to listed fish species. DWR has also treated the South Bay Aqueduct (SBA) to control algae that are stimulated by nutrient-rich Delta water. Including preventative treatments, DWR has treated the SBA for algal control between 10 and 16 times per year in recent years. Periodic treatment of the California Aqueduct and State Water Project (SWP) terminal reservoirs is also necessary for the same reason. Given the increasing environmental concerns about the use of copper-based algacides, it is likely that effective control will become increasingly more difficult and reduce the ability of downstream users to manage algae-related problems in the future.

Eutrophication, ecological stoichiometry and alternate stable state theories combine to serve as a unifying framework for understanding the complexity of responses not only in the Bay-Delta estuary but also, more generally, in many comparative systems. This interpretation does not negate the importance of ecological invasions, habitat changes, multiple stressors and food-web complexities, but adds an explanatory mechanism to those interpretations through biogeochemistry and organismal stoichiometry. Ecological stoichiometry affects systems by setting elemental constraints on the growth of organisms. This, in turn, affects food quality and the relationships between predators and prey.

Examples of Responses to Nutrient Load Reductions

A growing body of literature documents improvements in ecosystem functions where nutrient loading is reduced and stoichiometric balance is restored. Reducing nutrient loading in the Chesapeake Bay, Tampa Bay, and coastal areas of Denmark has proven to be effective at reversing the harmful effects of previously undertreated discharges and restoring the native food webs. For example, within several years of increasing nutrient removal at the Blue Plains treatment plant in Washington D.C., N:P ratios in the Potomac River declined, the abundance of the invasive *Hydrilla verticillata* and *Corbicula fluminea* began to decline (Figure 12 showing *Corbicula fluminea* and other relationships with N:P), and the abundance of native grasses increased (Ruhl and Rybicki 2010).

⁴ The reference condition is the 25th percentile of the nutrient data for sites within the ecoregion and is meant to represent the nutrient concentrations in minimally impacted water bodies.

⁵ Significant questions have been raised about the use of reference conditions to establish regulatory criteria for nutrients. Nevertheless, they provide a starting point for evaluating water quality.

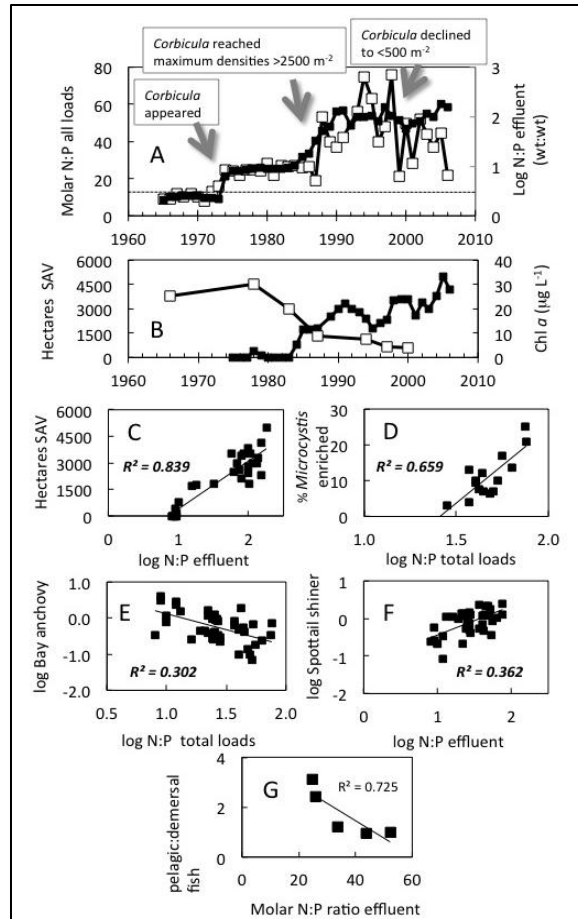


Figure 12. Comparative relationships for the Potomac River. Panel A shows the change in effluent N loading and the relative abundance of the invasive clam, *Corbicula fluminea*. *C. fluminea* appeared coincident with a sharp increase in N:P and increased in abundance as N:P increased. When N:P decreased sharply around 1999, *C. fluminea* abundance also declined sharply from $>2500\text{ m}^{-2}$ to $<500\text{ m}^{-2}$. Data derived from Dresler and Cory (1980), Jaworski et al. (2007), and Cummins et al. (2010). Figure reproduced from Glibert et al. (2011).

Tampa Bay provides another important example. Eutrophication problems in Tampa Bay were severe in the 1970s, with N loads approximating 24 tons per day, about half of which was due to point source effluent (Greening and Janicki 2006). Several years after nitrogen and phosphorus reductions were achieved, native seagrass began to increase. Lower nutrient discharges also had positive effects on the coastal waters around the island of Funen, Denmark (Rask et al. 1999). Since the mid 1980s, there has been a roughly 50% reduction in the loading of N and P in the region due to point source reductions. Again, native grasses returned and low oxygen problems were reversed.

Cloern (2001) provides additional examples of recovery following reductions in nutrient and waste inputs. Citing other researchers, Cloern (2001) shows improvements in dissolved oxygen levels in the Forth Estuary in Scotland following improvements in wastewater treatment. Citing a second study, Cloern (2001) shows increases in fish diversity in the Thames Estuary following improvements in wastewater treatment there (Figure 13).

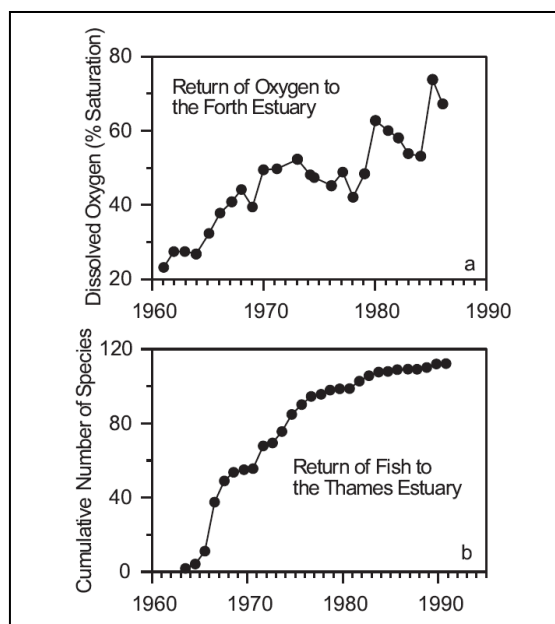


Figure 13. Two examples of recovery following actions to restore water quality in estuaries impacted by nutrient and waste inputs: (a) trend of increasing dissolved oxygen concentration (summer months) in the Forth Estuary, Scotland, following Improvements in wastewater treatment; (b) trend of increasing diversity of fishes in the Thames Estuary following implementation of advanced wastewater treatment and increases in oxygen concentrations (Source: Figure 20 from Cloern 2001).

Moreover, there is recent evidence that diatom blooms may be restored in the Bay-Delta estuary if NH_4 loading is reduced. In Suisun Bay, an unusual diatom bloom in Spring 2000 reached chl-*a* concentrations of $30 \mu\text{g L}^{-1}$ when NH_4 concentrations declined to $1.9 \mu\text{mol L}^{-1}$ (0.027 mg L^{-1}) (Wilkerson et al. 2006). Similarly, chl-*a* concentrations in Suisun Bay reached $35 \mu\text{g L}^{-1}$ during spring 2010 when NH_4 concentrations declined to $0.5 \mu\text{mol L}^{-1}$ (0.007 mg L^{-1}) (Dugdale et al. 2011, 2012). These blooms are comparable to spring chl-*a* levels from 1969 to 1977 (Ball and Arthur 1979) when NH_4 concentrations were $1.8 \mu\text{mol L}^{-1}$ (0.025 mg L^{-1}) during summer and $4.0 \mu\text{mol L}^{-1}$ (0.056 mg L^{-1}) during winter (Cloern and Cheng 1981).

In sum, ecological stoichiometry affects ecosystems by setting elemental constraints on the growth of organisms. However, as is the case with all conceptual frameworks, it must be viewed within the context of other factors in the environment, including the multiple stressors that now impact systems, as well as the scale (spatial, temporal and organismal) on which the system is being examined. Imbalances in stoichiometry may have impacts on ecosystems even at nutrient loads normally taken to be saturating or supersaturating. Ecological stoichiometry has several important implications for the health and sustainability of aquatic systems. Stoichiometric imbalances may accelerate transformations of nutrients or may alter the processes by which nutrients are cycled in the ecosystem, further altering nutrient availability or form for primary producers (Elser and Hamilton 2007, Nugraha et al. 2010). When food *quality* is linked to food web outcome, feedback effects and nutrient biogeochemical processes may play large roles in species success (Glibert 2012).

REFERENCES

- Ahlgren, G., L. Lundstedt, M. Brett, and C. Forsberg. 1990. Lipid composition and food quality of some freshwater phytoplankton for cladoceran zooplankters. *Journal of Plankton Research* 12: 809-818.
- Anderson, D. M., P. M. Glibert, and J. M. Burkholder. 2002. Harmful algal blooms and eutrophication: Nutrient sources, composition, and consequences. *Estuaries* 25: 704-726.
- Ball, M.D. and J. F. Arthur. 1979. Planktonic chlorophyll dynamics in the Northern San Francisco Bay and Delta. Pacific Division of the American Association for the Advancement of Science c/o California Academy of Sciences Golden Gate Park San Francisco, California 94118.
- Brett M.T. and D.C. Müller-Navarra. 1997. The role of highly un-saturated fatty acids in aquatic food-web processes. *Freshwater Biology* 38: 483-499.
- Britto DT, Kronzucker HJ (2002) NH_4^+ toxicity in higher plants: a critical review. *J Plant Physiol* 159: 567-584
- Cloern, J. E. 2001. Our evolving conceptual model of the coastal eutrophication problem. *Marine Ecological Progress Series* 210: 223-253.
- Cloern, J.E and R.T. Cheng. 1981. Simulation model of *Skeletonema costatum* population dynamics in Northern San Francisco Bay, California. *Estuarine, Coastal and Shelf Science*. 12:83-100
- Cloern, J.E. and R. Dufford. 2005. Phytoplankton community ecology: principles applied in San Francisco Bay. *Marine Ecology Progress Series*, 285:11-28.
- Coutteau P. and P. Sorgeloos. 1997. Manipulation of dietary lipids, fatty acids and vitamins in zooplankton cultures. *Freshwater Biology* 38: 501-512.
- Cummins, J., C. Buchanan, C. Haywood, H. Moltz, A. Griggs, R. C. Jones, R. Kraus, N. Hitt, and R. V. Bumgardner. 2010. Potomac Basin Large River Environmental Flow Needs. ICPRB Report 10- 3. Interstate Commission on the Potomac River Basin.
- Domingues, R.B., A.B. Barbosa, U. Sommer, and H.M. Galvao. 2011. Ammonium, nitrate and phytoplankton interactions in a freshwater tidal estuarine zone: potential effects of cultural eutrophication. *Aquatic Science* 73: 3331-343.
- Donald, D.B., M.J. Bogard, K. Finlay, L. Bunting and P.R. Leavitt. 2013. Phytoplankton-specific response to enrichment of phosphorus-rich surface waters with ammonium, nitrate, and urea. *PLoS One*, 8(1):e53277. Doi:10.1371/journal.pone.0053277.
- Dresler, P. V. and R. L. Cory. 1980. The Asiatic clam, *Corbicula fluminea* (Müller), in the tidal Potomac River, Maryland. *Estuaries*, 3: 150-152).
- Dugdale, R. C. and Goering, J. J. (1967). Uptake of new and regenerated forms of nitrogen in primary productivity. *Limnol. Oceanogr.* 12,196-206.
- Dugdale, R.C., F. P. Wilkerson, V. E. Hogue and A. Marchi. 2007. The role of ammonium and nitrate in spring bloom development in San Francisco Bay. *Estuarine, Coastal and Shelf Science*, 73: 17-29.
- Dugdale, R., F. Wilkerson, A. Parker. 2011. Brief report in response to selected issues raised by Sacramento Regional County Sanitation District in petition for review of discharge permit issued by the Central Valley Regional Water Quality Control Board. May 4, 2011.
- Dugdale, R.C., Wilkerson, F.P., and Parker, A.E. (2013). A biogeochemical model of phytoplankton productivity in an urban estuary: the importance of ammonium and freshwater flow. *Ecol.*

- Modeling* 263, 291-307.
- Dugdale, R.C., Wilkerson, F.P., Parker, A.E., Marchi, A., Taberski, K. 2012. River flow and ammonium discharge determine spring phytoplankton blooms in an urbanized estuary. *Estuarine and Coastal Shelf Science*. 115, 187-199
- Elser JJ, Hamilton A: Stoichiometry and the New Biology: The Future Is Now. *PLoS Biol* 2007, 5(7): e181. doi:10.1371/journal.pbio.0050181
- Eppley, R.W. and Peterson, B.J. (1979) Particulate organic flux and planktonic new production in the deep ocean. *Nature*, 282, 677–680.
- Færøvig, P.J. and D.O. Hessen. 2003. Allocation strategies in crustacean stoichiometry: the potential role of phosphorus in the limitation of reproduction. *Freshwater Biology* 48: 1782-1792.
- Feyrer F., B. Herbold, S.A. Matern, and P.B. Moyle. 2003. Dietary shifts in a stressed fish assemblage: Consequences of a bivalve invasion in the San Francisco Estuary. *Environmental Biology of Fishes* 67:277-288.
- Finkel Z.V., J. Beardall, K.J. Flynn, A. Quiqq, T.A. Rees, J.A. Raven. 2010. Phytoplankton in a changing world: Cells size and elemental stoichiometry. *J. Plankt. Res.* 2010, 32: 119-137.
- Flynn, K., J.M. Franco, P. Fernández, B. Reguera, M. Zepata, G. Wood, and K.J. Flynn, 1994. Changes in toxin content, biomass and pigments of the dinoflagellate *Alexandrium minutum* during nitrogen refeeding and growth into nitrogen and phosphorus stress. *Marine Ecological Progress Series* 111: 99-109.
- Foe, C., A. Ballard, and S. Fong. 2010. Nutrient Concentrations and Biological Effects in the Sacramento-San Joaquin Delta. Report prepared for the Central Valley Regional Water Quality Control Board.
- Gao, Y., J. Cornwell, D. Stoecker, and M. Owens. 2012. Effects of cyanobacterial-driven pH increases on sediment nutrient fluxes and coupled nitrification-denitrification in a shallow fresh water estuary. *Biogeosciences*, 9:2697-2710.
- Ger, K.A., S.J. Teh, D.V. Baxa, S. Lesmeister, and C.R. Goldman. 2010. The effects of dietary *Microcystis aeruginosa* and microcystin on the copepods of the upper San Francisco Estuary. *Freshwater Biology*, 55:1548-1559.
- Glibert, P.M. 1998. Interactions of top-down and bottom-up control in planktonic nitrogen cycling. *Hydrobiologia*. 363: 1-12.
- Glibert, P., 2010. Long-term changes in nutrient loading and stoichiometry and their relationships with changes in the food web and dominant pelagic fish species in the San Francisco Estuary, California. *Reviews in Fisheries Science*. 18(2):211-232.
- Glibert, P.M. 2012. Ecological stoichiometry and its implications for aquatic ecosystem sustainability. *Current Opinion Envir. Sustainability* 4:272-2.
- Glibert and Burkholder. 2011. Harmful algal blooms and eutrophication: Strategies for nutrient uptake and growth outside the Redfield comfort zone. *Chinese J. Limnol. Oceanogr.* 29: 724-738.
- Glibert, P.M., R.C. Dugdale, A.E. Parker, F. Wilkerson, J. Alexander, S. Blaser, E. Kress, and S. Murasko. 2012b. Elevated ammonium concentrations inhibit total nitrogen uptake and growth, not just nitrate uptake. Poster presentation at Interagency Ecological Program Annual Workshop, April 2012, Folsom, CA.

- Glibert, P., C.A. Heil, D. Hollander, M. Revilla, A. Hoare, J. Alexander, S. Murasko. 2004. Evidence for dissolved organic nitrogen and phosphorous uptake during a cyanobacterial bloom in Florida bay. *Mar Ecol Prog Ser*, 280:73-83.
- Glibert, P.M., D. Fullerton, J.M. Burkholder, J.C. Cornwell, and T.M. Kana. 2011. Ecological stoichiometry, biogeochemical cycling, invasive species, and aquatic food webs: San Francisco Estuary and comparative systems. *Reviews in Fisheries Science*, 19(4): 358-417.
- Glibert, P.M., T.M. Kana, K. Brown. 2013. From limitation to excess: the consequences of substrate excess and stoichiometry for phytoplankton physiology, trophodynamics and biogeochemistry, and the implications for modeling. *Journal of Marine Systems*, <http://dx.doi.org/10.1016/j.jmarsys.2012.10.004>.
- Glibert, P.M., F. Wilkerson, R.C. Dugdale, A.E. Parker, J.A. Alexander, S. Blaser and S. Murasko. In review. Microbial communities from San Francisco Bay Delta respond differently to oxidized and reduced nitrogen substrates – even under conditions that would otherwise suggest nitrogen sufficiency.
- Goebel, N.L., C.A. Edwards, J.E. Zehr, and M.J. Follows. 2010. An emergent community ecosystem model applied to the California Current System. *Journal of Marine Systems* 83: 211-242.
- Granéli E., and K. Flynn. 2006. Chemical and physical factors influencing toxin content. In: Granéli E, Turner, J.T. (Eds.), *Ecology of Harmful Algae*. Springer, Heidelberg, pp. 229-241.
- Greening, H. and A. Janicki. 2006. Toward reversal of eutrophic conditions in a subtropical estuary: Water quality and seagrass response to nitrogen loading reductions in Tamp Bay, Florida, USA. *Environ. Mgt.* 38(2):163-178.
- Ha, J.H., T. Hidaka, and H. Tsuno. 2009. Quantification of toxic *Microcystis* and evaluation of its dominance ratio in blooms using real-time PCR. *Envir. Sci. Technol.* 43: 812-818.
- Hall, E. K., F. Maixner, O. Franklin, H. Daims, A. Richter and T. Battin. Linking microbial and ecosystem ecology using ecological stoichiometry: A synthesis of conceptual and empirical approaches. *Ecosystems*, doi: 10.1007/s10021-010-9408-4 (2010).
- Harris, G.P. 1986. *Phytoplankton Ecology: Structure, Function and Fluctuation*. Cambridge University Press, Cambridge.
- Hecky, R.E. and P. Kilham. 1988. Nutrient limitation of phytoplankton in freshwater and marine environments: A review of recent evidence on the effects of enrichment. *Limnol. Oceanogr.* 33:796-822.
- Heil, C.A., Revilla, M., Glibert, P.M., Murasko, S., 2007. Nutrient quality drives phytoplankton community composition on the West Florida Shelf. *Limnol. Oceanogr.* 52, 1067-1078.
- Hendrixson, H. A., R. W. Sterner, and A. D. Kay. 2007. Elemental stoichiometry of freshwater fishes in relation to phylogeny, allometry and ecology. *Journal of Fish Biology* 70: 121-140.
- Hessen, D.O.. 1997. Stoichiometry in food webs – Lotka revisited. *Oikos* 79: 195-200.
- Hobbs, J.A., W.A. Bennett, and J.E. Burton. 2006. Assessing nursery habitat quality for native smelts (*Osmeridae*) in the low-salinity zone of the San Francisco estuary. *Journal of Fish Biology*, 69: 907–922
- Hood, J.M., and R.W. Sterner. 2010. Diet mixing: Do animals integrate growth or resources across temporal heterogeneity? *The American Naturalist* 176: 651-663.

- Ibanez, C., N. Prat, C. Duran, M. Pardos, A. Munne, R. Andreu, N. Caiola, N. Cid, H. Hampel, R. Sanchez, and R. Trobajo. 2008. Changes in dissolved nutrients in the lower Ebro river: Causes and consequences. *Limnetica*. 27(1):131-142.
- Jassby, A. 2008. Phytoplankton in the Upper San Francisco Estuary: recent biomass trends, their causes, and their trophic significance. *San Francisco Estuary and Watershed Science*. 6(1): Article 2, February 2008.
- Jassby, A.D., J.E. Cloern and B.E. Cole. 2002. Annual primary production: Patterns and mechanisms of change in a nutrient-rich tidal ecosystem. *Limnol. Oceanogr.*, 47(3): 698–712.
- Jaworski, N., W. Romano, and C. Buchanan. 2007. The Potomac River Basin and its estuary: Landscape loadings and water quality trends 1895-2005.
- Jeyasingh, P. D., and L.J. Weider. 2005. Phosphorus availability mediates plasticity in life-history traits and predator-prey interactions in *Daphnia*. *Ecology Letters* 8: 1021-1028.
- Jeyasingh, P. D. and L.J. Weider. 2007. Fundamental links between genes and elements: evolutionary implications of ecological stoichiometry. *Molecular Ecology* 16: 4649-4661.
- Johansson, N. and E. Granéli. 1999. Cell density, chemical composition and toxicity of *Chrysochromulina polylepis* (Haptophyta) in relation to different N:P supply ratios. *Marine Biology* 135: 209 - 217.
- Jordan, T. E., J. C. Cornwell, W. R. Boynton, and J. T. Anderson. 2008. Changes in phosphorus biogeochemistry along an estuarine salinity gradient: the iron conveyor belt. *Limnology and Oceanography* 53: 172-184.
- Kilham, S. S., D.A. Kreeger, C.E. Goulden, and S.G. Lynn. 1997. Effects of nutrient limitation on biochemical constituents of *Ankistrodesmus falcatus*. *Freshwater Biology* 38: 591-596.
- Kimmerer, W. J. (2004). Open water processes of the San Francisco Estuary: From physical forcing to biological responses. *San Francisco Estuary and Watershed Sci.*, 2. <escholarship.org/uc/item/9bp499mv>.
- Kimmerer W.J., A.E. Parker, U.E. Lidström, and E.J. Carpenter. 2012. Short-Term and Interannual Variability in Primary Production in the Low-Salinity Zone of the San Francisco Estuary. *Estuaries and Coasts* ,35:913–929.
- Kjørboe, T. 1989. Phytoplankton growth rate and nitrogen content: Implications for feeding and fecundity in a herbivorous copepod. *Marine Ecological Progress Series* 55: 229-234.
- Laspoumaderes, C., B. Modenutti and E. Balseiro. 2010. Herbivory versus omnivory: linking homeostasis and elemental imbalance in copepod development. *Journal of Plankton Research* 32: 1573-1582.
- Legendre, L. and F. Rassouzadegan. 1995. Plankton and nutrient dynamics in marine waters. *Ophelia* 41: 153-172.
- Lehman, P. W. (1996). Changes in chlorophyll-a concentration and phytoplankton community composition with water-year type in the upper San Francisco Estuary, in *San Francisco Bay: The Ecosystem*, ed. J. T. Hollibaugh (San Francisco: Pacific Division of the American Association for the Advancement of Science, 351-374) .
- Lehman, P. W. 2000. The influence of climate on phytoplankton community biomass in San Francisco Bay Estuary. *Limnol. Oceanogr.* 45: 580–590
- Lehman, P. W., G. Boyer, C. Hall, S. Waller and K. Gehrts. 2005. Distribution and toxicity of a new colonial *Microcystis aeruginosa* bloom in the San Francisco Bay Estuary, California. *Hydrobiologia*, 541:87-99

- Lehman, P.W., S.J. Teh, G.L. Boyer, M.L. Nobriga, E. Bass, and C. Hogle. 2010. Initial impacts of *Microcystis aeruginosa* blooms on the aquatic food web in the San Francisco Estuary. *Hydrobiologia*, 637:229-248.
- Lomas, M.W. and P.M. Glibert. 1999. Interactions between NH_4 and NO_3 uptake and assimilation: comparison of diatoms and dinoflagellates at several growth temperatures. *Marine Biology*, 133:541-551.
- Lopez, C.B., J.E. Cloern, T.S. Schraga, A.J. Little, L.V. Lucas, J.K. Thompson, and J.R. Burau. 2006. Ecological values of shallow-water habitats: implications for the restoration of disturbed ecosystems. *Ecosystems* 9: 422–440.
- McIntyre, P. B., and A. Flecker. 2010. Ecological stoichiometry as an integrative framework in stream fish ecology. *American Fisheries Society Symposium* 73: 539-558.
- Mitra, A. and K.J. Flynn. 2005. Predator-prey interactions: is “ecological stoichiometry” sufficient when good food goes bad? *Journal of Plankton Research* 27: 393-399.
- Moe, S. J., R.S. Stelzer, M.R. Forman, W.S. Harpole, T. Daufresne and T. Yoshida. 2005. Recent advances in ecological stoichiometry: insights for population and community ecology. *Oikos* 109: 29–39.
- Mousseau, L., B. Klein, L. Legendre, S. Dauchez, E. Tamigneaux, J.-E. Tremblay, and E.G. Ingram. 2001. Assessing the trophic pathways that dominate food webs: an approach based on simple ecological ratios. *J. Plank. Res.* 23: 765-777.
- Nielsen, K.J., 2003. Nutrient loading and consumers: Agents of change in open-coast macrophyte assemblages. *Proc. Nat. Acad. Sci.*, 100, 7660-7665.
- Nobriga, M.L. 2002. Larval delta smelt diet composition and feeding incidence: environmental and ontogenetic influences. *California Fish and Game* 88:149-164.
- Nobriga, M.L., F. Feyrer, R.D. Baxter, and M. Chotkowski. 2005. Fish community ecology in an altered river delta: spatial patterns in species composition, life history strategies, and biomass. *Estuaries*, 28(5):776-785.
- Nugraha, A, Pondaven P, Tréguer, P: Influence of consumer-driven nutrient recycling on primary production and the distribution of N and P in the ocean. *Biogeosci* 2010, 12: 1285-1305.
- Oh, H-M., S.J. Lee, M-H. Jang and B-D. Yoon. 2000. Microcystin production by *Microcystis aeruginosa* in a phosphorus-limited chemostat. *Appl. Envir. Microbiol.* 66: 176-179.
- Olsen, Y., 1999. Lipids and essential fatty acids in aquatic food webs: what can freshwater ecologists learn from mariculture? In: Arts, M.T., Wainman, B.C. (Eds). *Lipids in Freshwater Ecosystems*. Springer-Verlag, New York, pp. 161-202.
- Park, S., M.T. Brett, E.T. Oshel and C.R. Goldman. 2003. Seston food quality and *Daphnia* production efficiencies in an oligo-mesotrophic subalpine lake. *Aquatic Ecology* 37: 123-136.
- Parker, A.E., R.C. Dugdale, and F.P. Wilkerson. 2012a. Elevated ammonium concentrations from wastewater discharge depress primary productivity in the Sacramento River and the Northern San Francisco Estuary. *Marine Pollution Bulletin*. doi:10.1016/j.marpolbul.2011.12.016.
- Parker, A.E., V. E. Hogue, F.P. Wilkerson, and R.C. Dugdale. 2012b. The effect of inorganic nitrogen speciation on primary production in the San Francisco Estuary. *Estuarine, Coastal and Shelf Science*.
- Peñuelas, J., J. Sardans, A. Rivas-Ubach and I.A. Janssens. 2012. The human-induced imbalance between C, N and P in Earth’s life system. *Global Change Biology* 18: 3-6.

- Pilati, A. and M.J. Vanni. 2007. Ontogeny, diet shifts, and nutrient stoichiometry in fish. *Oikos* 116: 1663-167.
- Rask, N., S. E. Pedersen, and M. H. Jensen. 1999. Response to lowered nutrient discharges in the coastal waters around the island of Funen, Denmark. *Hydrobiologia* 393: 69–81.
- Redfield, A. C. On the proportions of organic derivatives in sea water and their relation to the composition of plankton, pp. 176-192. In: *James Johnstone Memorial Volume*. Liverpool: University of Liverpool Press (1934).
- Redfield, A. C. The biological control of chemical factors in the environment. *Am. Sci.*, 46: 205-221 (1958).
- Reynolds, C. S. 1984. Phytoplankton periodicity: the interactions of form, function and environmental variability. *Freshwater Biol.*, 14, 111–142.
- Ruhl, H.A. and N.B. Rybicki. 2010. Long-term reductions in anthropogenic nutrients link to improvements in Chesapeake Bay habitat. www.pnas.org/cgi/doi/10.1073/pnas.1003590107.
- Schindler, D. W. 1974. Eutrophication and Recovery in Experimental Lakes: Implications for Lake Management. *Science*. 184(4139):897-899.
- Slaughter, A. and W. Kimmerer. 2010. Abundance, composition, feeding, and reproductive rates of key copepod species in the food-limited Low Salinity Zone of the San Francisco Estuary. Poster Presentation at the 6th Biennial Bay-Delta Science Conference, Sacramento, CA, September 27-29, 2010.
- Sommer, T., C. Armor, R. Baxter, R. Breuer, L. Brown, M. Chotkowski, S. Culberson, F. Feyrer, M. Gingras, B. Herbold, W. Kimmerer, A. Mueller-Solger, M. Nobriga and K. Souza. 2007. The collapse of pelagic fishes in the upper San Francisco Estuary. *Fisheries*, 32(6):270-277.
- Sterner, R.W. and J.J. Elser. 2002. *Ecological stoichiometry: The biology of elements from molecules to the biosphere*. Princeton University Press, Princeton, N.J.
- Sterner, R. W. and N.B. George. 2000. Carbon, nitrogen, and phosphorus stoichiometry of cyprinid fishes. *Ecology*, 81, 127-140.
- Teh, S., I. Flores, M. Kawaguchi, S. Lesmeister and C. Teh. 2011. Full Life-Cycle Bioassay Approach to Assess Chronic Exposure of *Pseudodiaptomus forbesi* to Ammonia/Ammonium. Report Submitted to Chris Foe and Mark Gowdy at the State Water Resources Control Board, August 31, 2011.
- U.S. Environmental Protection Agency. 1980. Trophic State of Lakes and Reservoirs, Technical Report E-80-3.
- U.S. Environmental Protection Agency. 2001a. Ambient Water Quality Criteria Recommendations: Information Supporting the Development of State and Tribal Nutrient Criteria; Rivers and Streams in Ecoregion I. U.S. Environmental Protection Agency, Office of Water, EPA 822-B-01-012 (December 2001).
http://www.epa.gov/waterscience/criteria/nutrient/ecoregions/rivers/rivers_1.pdf
- U.S. Environmental Protection Agency. 2001b. Ambient Water Quality Criteria Recommendations: Rivers and Streams in Ecoregion I. U.S. Environmental Protection Agency, Office of Water (December 2001).
- U.S. Environmental Protection Agency. 2012. Water Quality Challenges in the San Francisco Bay/Sacramento-San Joaquin Delta Estuary: EPA's Action Plan. [Available at <http://www.epa.gov/sfbay-delta/pdfs/EPA-bayareaactionplan.pdf>]

- U.S. Environmental Protection Agency. 2013. Aquatic Life Ambient Water Quality Criteria for Ammonia – Freshwater 2013. EPA 822-R-13-001. April 2013. Available at <http://water.epa.gov/scitech/swguidance/standards/criteria/aqlife/ammonia/upload/AQUATIC-LIFE-AMBIENT-WATER-QUALITY-CRITERIA-FOR-AMMONIA-FRESHWATER-2013.pdf>
- U.S. Fish and Wildlife Service. 2012. Endangered and Threatened Wildlife and Plants; 12-month Finding on a Petition to List the San Francisco Bay-Delta Population of the Longfin Smelt as Endangered or Threatened. 50 CFR Part 17, Docket No. FWS-R8-ES-2008-0045, 4500030113. 77 Fed. Reg. 19755, 19776.
- Van de Waal, D. B., J. M. Verspagen, M. Lurling, E. Van Donk, P. M. Visser and J. Huisman. 2009. The ecological stoichiometry of toxins produced by harmful cyanobacteria: An experimental test of the carbon-nutrient balance hypothesis. *Ecology Letters* 12: 1326-1335.
- Van Nieuwenhuysse, E. 2007. Response of summer chlorophyll concentration to reduced total phosphorus concentration in the Rhine River (Netherlands) and the Sacramento-San Joaquin Delta (California, USA). *Canadian Journal of Fisheries and Aquatic Sciences* 64: 1529-1542.
- Walve, J. and U. Larsson. 1999. Carbon, nitrogen and phosphorus stoichiometry of crustacean zooplankton in the Baltic Sea: implications for nutrient recycling. *Journal of Plankton Research* 21: 2309-2321.
- Ware, D. M., and R. E. Thompson. 2005. Bottom-up ecosystem trophic dynamics determine fish production in the Northeast Pacific. *Science*: 308: 1280-1284.
- Weers, P.M.M. and R.M. Gulati. 1997. Effects of the addition of polyunsaturated fatty acids to the diet on the growth and fecundity of *Daphnia galeata*. *Freshwater Biology*: 38, 721-729.
- Wilkerson, F.P., R.C. Dugdale, V.E. Hogue and A. Marchi. 2006. Phytoplankton blooms and nitrogen productivity in San Francisco Bay. *Estuaries and Coasts*, 29(3): 401–416.
- Wilkerson, F.P., A.E. Parker, and R.C. Dugdale. (in preparation). Application of enclosure experiments to characterize potential phytoplankton productivity in rivers and estuaries.
- Winder, M. and A.D. Jassby. 2010. Shifts in zooplankton community structure: Implications for food web processes in the Upper San Francisco Estuary. *Estuaries and Coasts*. DOI 10.1007/s12237-010-9342-x.
- Yamamoto, T. 2002. The Seto Inland Sea—eutrophic or oligotrophic? *Marine Pollution Bulletin* 47(1): 37 – 42.
- Yarrow, M., V. H. Marin, M. Finlayson, A. Tironi, L. E. Delgado, and F. Fischer. 2009. The ecology of *Egeria densa* Planchón (Liliopsida: Alismatales): A wetland ecosystem engineer? *Revista Chilena de Historia Natural* 82: 299-313.
- Yoshiyama, K. and J.H. Sharp. 2006. Phytoplankton response to nutrient enrichment in an urbanized estuary: apparent inhibition of primary production by overeutrophication. *Limnology and Oceanography* 51: 424-434.